SEDIMENT OXYGEN DEMAND AND BIOCHEMICAL OXYGEN DEMAND: PATTERNS OF OXYGEN DEPLETION IN TIDAL CREEK SITES

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ABSTRACT

Concerns about low dissolved oxygen levels in New Hanover County, North Carolina tidal creeks resulted in a study measuring rates of oxygen loss as water-column biochemical oxygen demand (BOD_5) and sediment oxygen demand (SOD). Selected tidal creek sites were sampled monthly from July 2001 to August 2002 in collaboration with the New Hanover County Tidal Creeks monitoring project to identify any trends that may be evident concerning rates of oxygen loss. BOD_5 rates ranged from 0.0 to 7.6 mg l^{-1} and were strongly correlated with chlorophyll *a* measurements. This indicates that conditions leading to algal blooms have the potential to cause increased BOD and thus contribute to hypoxia in tidal creeks. SOD rates ranged from –1.5 to 6.3 g O_2 m^{-2} d^{-1}. Both rates of oxygen uptake were seasonally dependent. Oxygen loss to sediments was greater and more variable than oxygen loss in the water column, indicating that SOD should be considered in all comprehensive water quality monitoring programs. Results indicate that sediment composition and the bioavailability of organic matter may be key elements in determining SOD. On-land processes that increase sedimentation of organic material may contribute to creek hypoxia incidents. Groundwater inputs decreased SOD rates at selected sites, thus recharge areas may be critical to tidal creek health. Correlation and principal component analyses were conducted using SAS statistical software to assess the effects of numerous variables on oxygen demand. The results suggest that BOD_5 responds to a suite of environmental variables including temperature, salinity, chlorophyll *a*, total and organic suspended solids and rainfall while SOD responds to salinity and nutrients.

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DEDICATION

I would like to dedicate this work to my grandparents Dorothy and Paul Faherty, the two most beautiful, loving and kind people I have ever known. Without them in my life I would have never known how wonderful the world can be. Thank you for sharing so much with me and for helping me to become who I am. Although you are not here to read this, I know that you are in my heart and dreams.

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INTRODUCTION

Tidal creeks along the coast of the southeastern United States periodically experience low dissolved oxygen levels, possibly related to anthropogenic inputs. Some creeks also host periodic algal blooms (Mallin et al., 2003), which likely affect oxygen demand rates. The presence of adequate amounts of dissolved oxygen (DO) in aquatic environments is critical for maintaining high quality water and healthy populations of organisms, including fish. The North Carolina water column dissolved oxygen standard is 5 mg l^{-1}. Levels have been found well below this in North Carolina (Mallin et al., 2000, 2003) and South Carolina tidal creeks (Lerberg et al., 2000).

Dissolved oxygen (DO) concentrations are a general indicator of the overall health of a creek system. This is due to the low solubility of oxygen in water compared to other gases, such as CO_2. DO measurements provide information about biochemical and biological reactions occurring in water. Oxygen concentrations in bottom waters and interactions between nutrients, metals and sediments are heavily influenced by oxygen consumption or uptake (Orren, 1999).

The effects of hypoxia (<2 mg l^{-1} of oxygen) and anoxia (a complete lack of oxygen) can ultimately affect aquatic ecology and human populations though loss of resources and water pollution. Poor sanitation and sewage disposal practices have led to major human health epidemics including typhoid, cholera and dysentery in the past (Davis, 1950). BOD and SOD rates can be measures of poor sewage disposal practices or other sources of pollution.

Background

Dissolved Oxygen

Loss of oxygen in aquatic systems occurs during water column decomposition of organic matter and oxygen uptake into sediments. Decay of organic matter by bacteria, ammonia oxidation by nitrifiers, algal respiration and flux of oxygen into the sediment all increase oxygen demand (Walker and Snodgrass, 1986). As temperatures increase so does the demand for oxygen due to an increase in respiration in the biological community (Laws, 1993). Increased respiration is coupled with a decrease in the capacity of water to dissolve oxygen under warm conditions (Laws, 1993). The most common occurrences of hypoxia and anoxia are in the summer when waters may be stratified and temperatures are higher.

Extreme oxygen consumption, which occurs when oxygen is not replaced by aeration or primary productivity, may create anoxic conditions that result in fish kills, invertebrate die offs, species displacement and water quality decline. Low D.O. levels (1.5-2 mg l^{-1}) may also lead to displacement of some invertebrates and fish species due to loss of useable habitat (Levings, 1980). Anoxic waters can be fatal for shellfish and other sessile organisms since these organisms cannot relocate themselves to a more oxygenated area. Hypoxic and anoxic bottom waters often cause community shifts from larger longer lived to smaller shorter-lived species, due to differences in sensitivity to oxygen depletion (National Research Council, 2000).

There are numerous chemical effects that occur when there are low dissolved oxygen levels in coastal systems (Orren 1999). When waters become hypoxic, nitrate converts to nitrite, changing the available inorganic form of dissolved nitrogen. Some denitrification occurs releasing nitrogen and nitrous oxide gas. Since most organisms cannot assimilate nitrogen gas as a nutrient source this results in a net loss of bioavailable nitrogen from a system (An and Joye, 2001). With additional oxygen reduction insoluble metal oxides begin to become solubilized as an alternative form of dissolved oxygen. First manganese is reduced and then iron oxides. Finally, sulphate ions may become anaerobically converted to highly toxic sulfides by sulfate-reducing bacteria. Trace elements that may be toxic to organisms also become a greater threat when DO levels are low (Orren, 1999). When precipitated oxides re-dissolve loads of highly toxic trace elements are released, which may include mercury, lead and adsorbed nutrients. In combination, other reduced organic and inorganic compounds may also be toxic to benthic organisms. Thus, it is important to continuously monitor the ambient DO levels and DO uptake levels in the water column and the sediment surface in areas where water quality is of concern.

Biochemical Oxygen Demand

The oxygen consumption of the water column is measured as Biochemical Oxygen Demand (BOD) given in mg l^{-1}. BOD is defined as biochemical oxygen demand because the decline in oxygen is from a combination of chemical and biological processes.

This is the sum of carbonaceous and nitrogenous demand. BOD is a measure of the molecular oxygen utilized during a specific incubation period (Five or twenty five days to give BOD_5 or BOD_{20}) for the biochemical degradation of organic material (carbonaceous demand) and the oxygen used to oxidize inorganic material (nitrogenous demand) as well as the amount of oxygen used to reduce forms of nitrogen (Eaton et al., 1995).

Sediment Oxygen Demand

Sediment oxygen demand (SOD) is comprised of biological sediment oxygen demand (BSOD) and chemical sediment oxygen demand (CSOD), measured as g O_2 m^{-2} d^{-1}. SOD creates oxygen deficits in water bodies by reducing the amount of available oxygen in the water column (Hatcher, 1950; Seiki et al., 1994). SOD can be a significant percentage of the total oxygen uptake in aquatic systems (Caldwell and Doyle, 1995, Rounds and Doyle, 1997). Measurements of SOD give indications about decomposition rates of settling detritus and regeneration rates of nutrients from the sediment (remineralization) (Seiki et al., 1994). SOD rates also serve as proxies for the effects of pollution and other environmental factors on the biological activity of the benthic community. A nutrient loaded system often has an increased demand for oxygen (Natural Research Council, 2000).

BSOD is dominated by aerobic heterotrophic bacteria that utilize organic material as an energy source for growth (Walker and Snodgrass, 1986) and tends to occur at the sediment surface. Benthic oxygen uptake rates may be a function of the velocity of the overlying water. If there is a lack of water movement oxygen gradients may become very unstable due to local distortion by faunal activity that causes oxygen fluctuations in the water above the sediment (Revsbech et al., 1980).

It is thought that the rate-limiting step for oxygen flux into the sediments is through transport within sediments rather than through transport in the water column. Walker and Snodgrass (1986), however, found that aerobic oxidation relies more on the biodegradability or the flux of organic matter to the sediments than the quantity of organics present.

CSOD is a reaction that occurs deeper in the sediment (several centimeters) in an anoxic-anaerobic region where anaerobic bacteria degrade organic matter (Rounds and Doyle, 1997). This process produces reduced ions that react with oxygen when they diffuse upward to an oxidized zone (Walker and Snodgrass, 1986). In some water bodies it has been shown that biological consumption may control SOD in summer and fall while chemical consumption may be dominant in winter and spring (Seiki et al., 1994).

Sediment oxygen demand may be related to numerous variables. These include temperature, water column dissolved oxygen concentrations, organic matter content, sediment grain size, flow rate, sediment disturbance, toxic substances and measurement techniques (Medine et al., 1980; Krantzberg, 1994).

Temperature has often proven to be significantly correlated with SOD rates. SOD rates linearly increase with temperature (Seiki et al., 1994, Hu et al., 2001). Rates in summer and fall are often reported to be higher than those in winter. This increase with temperature may be partly due to increases in biological oxygen consumption resulting from a shift in bacterial species that have different optimal temperatures for growth rate (Walker and Snodgrass 1950).

Low dissolved oxygen levels in the water column (below 2-3 mg l^{-1}) may negatively influence SOD rates by limiting the supply of DO. SOD does not appear to be affected at higher oxygen concentrations (Jensen and Adrian, 1989). Decomposing algal detritus from large algal populations may be a factor that lowers dissolved oxygen levels locally. This is not a primary source of organic matter for SOD because most comes from non-algal sources (Rounds and Doyle, 1997). Extreme levels of SOD (19.5 g m^{-2} d^{-1}) may be partly due to oxygen consumption by settled sewage material (Davis, 1950).

Some studies have found that sediment type influences sediment oxygen demand, with sediments rich in organics consuming the most oxygen, but other studies found no correlation (Seiki et al., 1994). Minute amounts of biodegradable organics that exist in sandy environments can also efficiently consume oxygen (Seiki et al., 1994). SOD rates around 1-4 g m^{-2} d^{-1}, typical of sandy sediments with little organic matter, are still high enough to be important oxygen sinks (Rounds and Doyle, 1997).

The velocity of water movement in the water column is another variable that may affect SOD rates (Whittemore, 1950). Flow may play a role in determining diffusive fluxes and concentration gradients. The thickness of the near-bottom boundary layer, which is dependent on flow rates (Vogel, 1981), affects near-bottom concentration gradients of dissolved gases and nutrients (Cahoon, 1988). In the absence of flow, oxygen gradients may become unstable. SOD rates may be higher during low flow periods, partly due to increased deposition (Hatcher, 1950). A study conducted by Mackenthun and Stefan (1998) concluded that SOD rates are less dependent on flow when the upper sediment layers are depleted of organics. SOD rates in tidal creeks that have drastic flow and salinity changes throughout a tidal cycle may be affected differently.

Polluted water bodies, such as bays, with high sediment oxygen demand (due to high amounts of labile organic material) often are not suitable for colonization by benthic invertebrates (Krantzberg, 1994). In these areas, improvement of dissolved oxygen levels through activities such as dredging are crucial to maintaining healthy benthic populations and reducing metal bioavailability. Suspended sediment related oxygen depletion may also occur, however (Lee and Jones, 1999).

There are several ways of measuring sediment oxygen demand. The most common methods use intact cores or *in situ* bell chambers. The coring method is often used in deeper waters or for manipulation experiments in the laboratory. Intact cores are collected in the field and undisturbed layers are brought to the laboratory for analysis using a respirometer or other method after incubation (Seiki et al., 1994; Shin et al., 2000). Although this method is commonly employed, core handling and compaction may alter the biological community of the core and the metabolic processes occurring within the core, affecting SOD rates (Murphy and Hicks, 1950).

SOD chambers can be secured to the bottom sediment for *in situ* measurements (Caldwell and Doyle, 1995; Rounds and Doyle, 1997). Benthic chambers allow for measurement of the loss of dissolved oxygen in a known volume of water. Enclosed water may be analyzed using Winkler Titrations (Strickland and Parsons, 1972) or oxygen electrodes that are inserted into the chambers to measure dissolved oxygen concentrations over time. These *in situ* measurements produce representative SOD rates in the chambers after a two-hour time period (Caldwell and Doyle, 1995). The accuracy of these measurements relies on the ability of the chamber to simulate natural conditions (Boynton et al., 1981), including temperature and flow.

Although methods of measurement such as intact cores brought to the laboratory and *in situ* bell chambers give results that are comparable (Seiki et al., 1994), an *in situ* method gives the best estimates of SOD, due to minimal manipulation of sediments (Murphy and Hicks; 1950, Cahoon, 1988), sampling of a larger sediment area (Cahoon, 1988) and other environmental factors such as maintaining natural temperature, water flow and physical and chemical properties of the water column.

Study Objectives

Although the water quality of local tidal creeks had been studied, as part of the New Hanover County Tidal Creeks program by UNCW/Center for Marine Science's Aquatic Ecology Laboratory, for seven years a seasonal study of biochemical and sediment oxygen demand had never been undertaken in these areas. Due to the different intensities of residential development and other types of biological and geological variables in the creek watersheds these areas serve as a natural laboratory for the study of numerous impacts on water quality. Oxygen demand studies may reveal important seasonal and spatial oxygen depletion patterns in these sites that can help explain low dissolved oxygen levels.

The rate of oxygen removal by aquatic sediments is important in determining the response of water bodies to wastewater discharge and water quality management (Medine et al. 1980). Removal rates must be known in order to manage a water body for dissolved oxygen through discharge permit specifications and to avoid water quality impacts (Hatcher, 1950). Oxygen demand measurements should be considered in all comprehensive water quality monitoring programs. Results from this research may provide oxygen demand data that will assist in determining proper management techniques that can be employed to assist in protecting New Hanover County watersheds.

Hypotheses addressed in this study include:

H_1: SOD is a significant portion of the total oxygen demand in the chosen tidal creek study sites.

H_2: SOD and BOD_5 rates are seasonally dependent in chosen study sites

H_3: High SOD rates are correlated with organic matter or grain size content in bottom sediments of study sites.

The six main objectives were to:

1) Determine monthly water column BOD_5 levels at selected sites in Futch, Hewletts and Pages Creeks.

2) Determine monthly SOD rates in tidal creek sites.

3) Determine physical parameters and rates of water flow at study sites.

4) Determine the monthly total and organic suspended solid concentrations in study sites.

5) Determine seasonal organic content, grain size and carbohydrates (total and soluble) of bottom sediments of study sites.

6) Establish BOD_5 and SOD rates and correlate them to variables measured.

METHODS

Incubations were conducted to determine seasonal biochemical oxygen demand (measured as BOD_5) and sediment oxygen demand (SOD) rates in tidal creek environments within New Hanover County, North Carolina from July, 2001-August, 2002. Incubations were conducted once a month during the summer, fall, winter and spring seasons, in collaboration with the New Hanover County Tidal Creeks project, to identify any trends concerning BOD_5 and SOD rates. Sediment oxygen demand may be related to numerous variables, thus percent organic content, total and soluble carbohydrate content, grain size, total suspended solids of sediments, and water flow were measured at each study site.

Study Sites

Study sites were chosen in Futch Creek, Pages Creek and Hewletts Creek (Fig. 1) based on general water quality parameters such as dissolved oxygen and nutrient loading.

Futch Creek is a tidal creek with few algal blooms and low fecal coliform levels (Mallin et al., 1998). Numerous natural springs feed into this creek. The upper southern branch has shallow surface feeder creeks (< 30 cm deep at low tide) and a small upstream spring (Mallin et al., 1996). The mouth of this creek was dredged in 1995-1996 to improve microbiological water quality and lower portions of the creek have been re-opened to shell fishing (Mallin et al., 2000). Tidal Creek water quality reports indicate that periodic low summer dissolved oxygen levels creek-wide are the only water quality

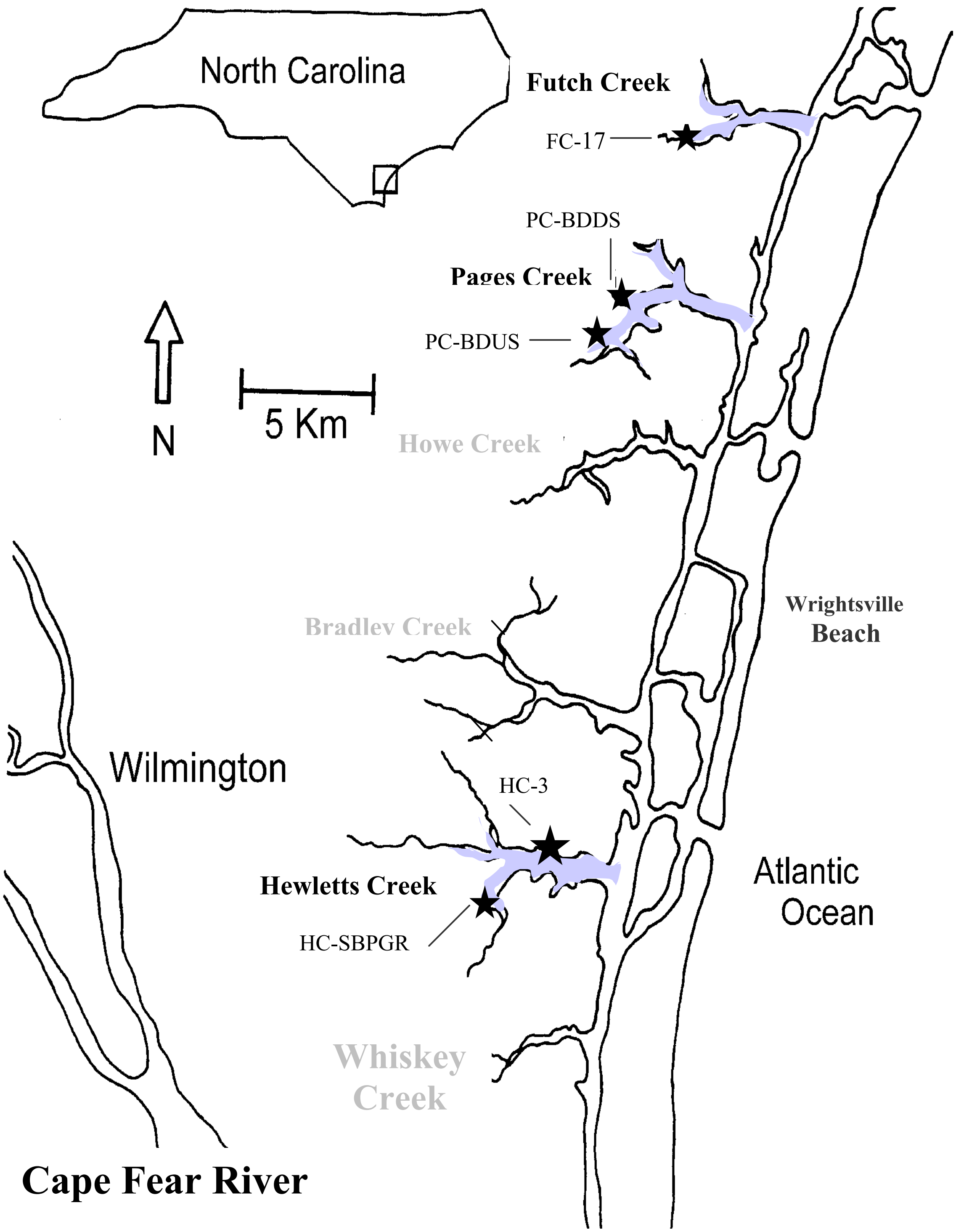

Figure 1.
Coastal Wilmington and New Hanover County, NC Tidal Creeks
(Mallin et al., 2000)
★ indicates study sites.

problem in this creek. The chosen study site on this creek (FC-17) lies adjacent to a residential dock in the upper south branch of the creek (Fig. 2a). Numerous study sites were inaccessible on this creek, thus only one study site was chosen for this research.

Pages Creek is not highly impacted by nutrient loading. Study sites were located in areas that experience the highest levels of fecal coliforms, chlorophyll *a* and sediment metals in the entire creek (Mallin et al., 2000). The study site PC-BDUS, in the upper branches of Pages Creek, has a spring visibly bubbling up at low tide. Both of the sites chosen (PC-BDDS and PC-BDUS) have experienced periodic hypoxia in the summer months and rare algal blooms (Mallin et al., 1998). The downstream site is adjacent to a private residential dock and the upstream site is adjacent to a boat dock (near a natural spring) (Fig. 2b). The upper branches of Pages Creek have experienced high sedimentation and in filling due to runoff and development.

The third creek examined, Hewletts Creek, receives high nutrient loading, experiences periodic algal blooms and has low dissolved oxygen levels in warmer months (Mallin et al., 2003). Portions of the creek run along roadways and collect run-off from golf courses and suburban areas. Nitrate concentrations have been high in portions of the creek (Mallin et al., 2003). The presence of non-toxic forms of *Pfiesteria piscicidia* were confirmed in phytoplankton samples at the south branch study site in the summer of 1995 (Mallin et al., 1998). Study sites in this creek were HC-3, a main tributary site adjacent to a private dock in the main channel of the creek and HC-SBPGR, adjacent to Pine Grove Road (Fig. 2c).

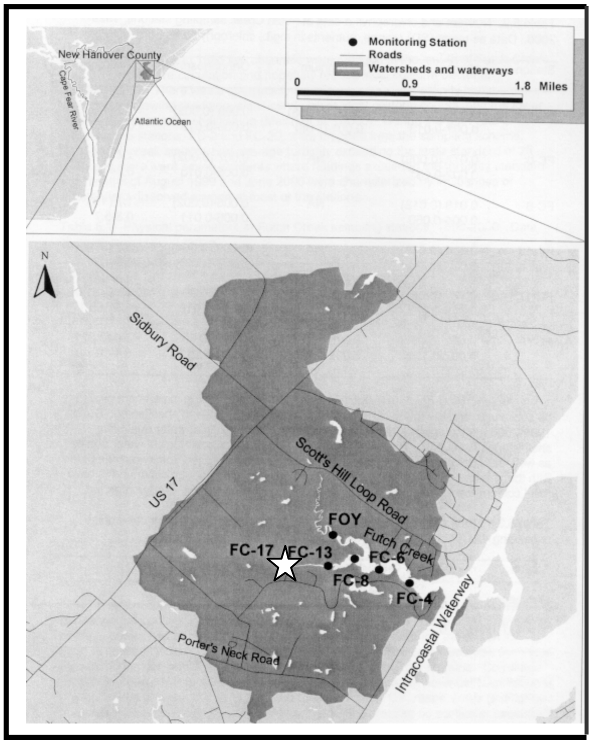

Figure 2a. Futch Creek Watershed
☆ = study site
(Mallin et al., 2000)

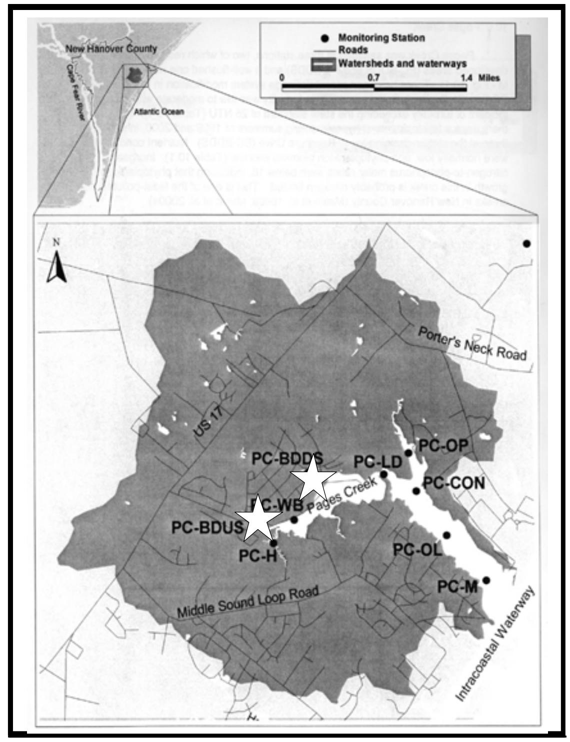

Figure 2b. Pages Creek Watershed
☆ = study sites
(Mallin et al., 2000)

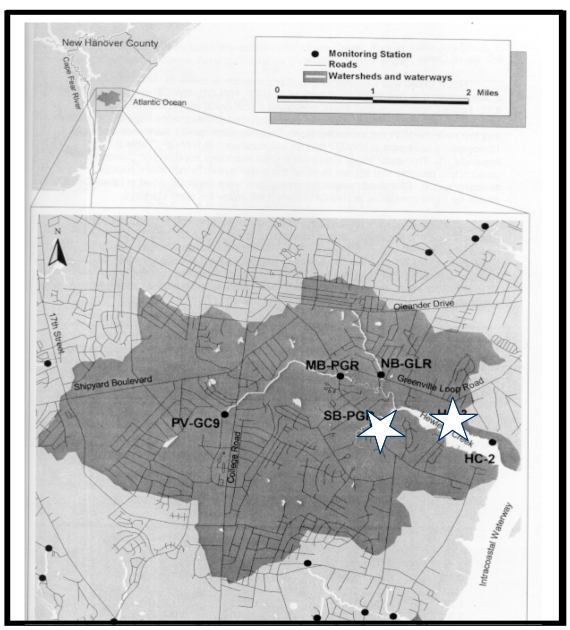

Figure 2c. Hewletts Creek Watershed
☆ = study sites
(Mallin et al., 2000)

Field Methods

Dissolved oxygen (DO), temperature, salinity and conductivity data were collected with a YSI Model 85 meter before BOD_5 and SOD samples were taken. Nutrient and chlorophyll *a* data were obtained from the Aquatic Ecology Laboratory for all sites. These measurements were taken monthly as part of a water quality monitoring program on the same day that oxygen demand data were obtained. Flow measurements were taken using a Marsh-McBirney Flo-Mate Model 2000 flow meter with precision of $\pm$ 2% and a range of -0.15 to 6 m sec^{-1}.

Water column biochemical oxygen demand (BOD_5) samples were taken simultaneously with triplicate SOD measurements. Samples were collected in the field with one-liter Nalgene™ bottles for duplicate analysis at all study sites. Collection bottles were put on ice, kept in the dark and brought to the laboratory for analysis within six hours.

SOD measurements were made using triplicate SOD chambers that were set out at each study site, on the rising tide, at the sediment water interface. Chambers were made of 20 cm diameter" PVC pipe with beveled edges on the bottom for easy entry into the sediment, as described by Cahoon (1996) (Fig. 3). Clear 0.5 cm diameter tubing inserted into the side of the chamber allowed for extraction of samples (Fig. 4). Each chamber had a plastic spout that was plugged with a rubber stopper once initial samples were taken. The chambers were placed in enough water to ensure that the natural flow of the system was maintained. This was done through the use of whirling cup rotors above the chambers and propellers within the chambers (Cahoon, 1988) set in motion by

Figure 3. Sediment oxygen chamber with external dimensions: 26x16 cm. (Cahoon, 1996).

Figure 4. Sample extraction in the field.

the flow of the creek. Rotors and propellers ensured uniform DO concentrations within the chamber at all times. Initial and final samples were extracted from each chamber into 60 ml dissolved oxygen bottles using large syringes. Once initial samples were taken the chambers were plugged so that there was no further introduction of water. A two- hour time span was allowed between extraction of initial and final samples to ensure that a rate for oxygen demand was established within the chambers (Rounds and Doyle, 1997). Collected samples were kept on ice and brought back to the laboratory for analysis within forty-eight hours.

Sediment samples were collected in the water column and in creek beds. Duplicate one-liter water samples were collected at study sites each month for total and organic suspended solid determination. Sediment cores (10 cm and "fluff layer") were taken using PVC pipe with an 8 cm diameter. For the purposes of this study "fluff layer" was defined as the top few centimeters that, when cored, were different in color and texture that the remaining cores sediment. Sediment core samples were frozen for later analysis.

Laboratory Analysis

Biochemical Oxygen Demand

BOD_5 was reported as the amount of oxygen consumed in a 300 ml sample of water incubated in the dark at 20° Celsius for 5 days (Laws, 1993). Incubations were conducted in the dark to prevent the photosynthetic production of DO. Samples were processed using the 5-day BOD test described by Eaton et al. (1995). (There was a linear regression for BOD uptake over five days in study sites with high and low phytoplankton abundance). All dissolved oxygen measurements were made in mg l^{-1} with an YSI Model 57 dissolved oxygen meter. BOD_5 rates were converted to g O_2 m^{-2} d^{-1} using the following equation:

(1) $\mathbf{BOD\ g\ O_2\ m^{-2}\ d^{-1} = (BOD_5 \times IWD)/\ 5}$

IWD = initial water depth when chambers were placed (m)

5= five day BOD incubation

Sediment Oxygen Demand

Laboratory analysis of triplicate water samples from SOD chambers were conducted within forty- eight hours of collection using Winkler Titrations (Strickland and Parsons, 1972) in order to establish a rate of oxygen demand over time. Samples were warmed to room temperature before analysis. In order to determine rates of SOD the following equations were used (Strickland and Parsons, 1972):

To determine mmO_2 l^{-1} of water in the chambers:

(2) $\mathbf{mg\ O_2\ l^{-1} = S/(S - 0.4) \times (5/50) \times f \times T \times 16}$

S= volume of SOD bottle

0.4= reagent volume added to fix samples in the field

5ml= 0.01 iodate solution volume used in calibration constant

50ml= aliquot taken from sample bottle

f-value= thiosulphate calibration constant determined monthly

T= volume of thiosulphate titrated

16= factor to equate mm O_2 l^{-1} to mg O_2 l^{-1}

Rates of demand will be reported as g O_2 m^{-2} d^{-1}. The final rate of SOD is calculated as:

(3) $$\textbf{SOD (g } \mathbf{O_2\ m^{-2}\ d^{-1}}\textbf{)=((F-I) x (V/A)) x 2 } \mathbf{h^{-1}}\textbf{x } \mathbf{1000^{-1}}\textbf{ x 24 h}$$

F-I= final- initial concentration in mg O_2

V= 4.49 L (volume of water in chamber)

A= 0.02138 m^2 (area of chamber)

2h= amount of incubation time in the field

1000= factor to convert mg units to g

Individual rates were calculated for each of the three chambers at each study site.

Sediment Analysis

Total and organic suspended solids (TSS and OSS) were determined monthly by filtering duplicate recorded volumes of sample water (~400 ml) through pre-weighed A/E Gelman 47 mm diameter glass fiber filters. Samples were placed in a drying oven for 24 hours at 60°and then reweighed. To determine TSS (mg l^{-1}) the initial weight of the filter paper was subtracted from the dried sediment weight and divided by the volume of water filtered. Samples were then combusted at 400° for four hours to determine organic and inorganic suspended solids by loss of organic matter on ignition.

Organic content, grain size and carbohydrate content of sediment cores were analyzed in July and October (2001) and January and May (2002). Percent organic content of sediments was determined using 10 cm sediment cores and various fluff layer samples from each study site. A well-mixed sub-sample of thawed sediment was weighed in aluminum pans and dried overnight at 60 °C. Dry weights were obtained before samples were combusted at 450 ^{o}C for four hours. Finally, inorganic weights were used to determine the percent organic content of sediment.

Grain size analysis was conducted using thawed sediment cores. Sediment was heated and treated with a 30 % hydrogen peroxide solution to oxidize organic matter before grain size analysis (Folk, 1980). Samples were then well mixed and analyzed with a LS 230 Beckman Coulter Particle Sizer.

A carbohydrate analysis was conducted to determine the seasonal total and soluble organic content of bottom sediments at all study sites using the methods of Underwood et al., (1995). A small amount of freeze-dried sediment (~5mg) was weighed in a glass tube and 1 ml of 25% saline solution was added. Samples were vortexed and reacted with 0.5 ml 10% phenol solution and 2.5 ml of concentrated sulphuric acid to give a vigorous exothermic reaction. (Soluble carbohydrate samples were extracted and decanted before this step took place). An incubation period of 45 minutes was allowed for all samples including blanks and standards to ensure proper color development. The reaction solution was decanted to a centrifuge tube for a fifteen-minute centrifugation at 2500 rpm. The supernatant was then poured into cuvettes and absorbance was measured on a Bausch& Lomb Spectronic 400 at 485 nm. Standard were made use glucose and results are expressed as glucose equivalents. Absorbance values were used to calculate total/soluble carbohydrates using the following formula:

$$(4)\quad \text{Carbohydrate } (\mu g/g) = \frac{(\text{Abs. Sample} - \text{Abs. Blank}) / (\text{Slope of Standard Curve})}{\text{Initial Sample Weight (g)}}$$

Statistical Analysis

Data formatting was done using Microsoft Excel™. Normality tests and Pearson correlation analyses were conducted for all variables using SAS statistical software (1998). Variables that were not normally distributed were log transformed ($\log_{10} +1$). An ANOVA test (SAS) was conducted using BOD_5 data to determine statistical significance between sites. Kruskal-Wallis and Wilcoxon 2-sample nonparametric tests (SAS) were conducted using SOD data to determine levels of significance where differences existed between individual study sites. Finally, a principal component analyses was conducted using SAS. This analysis divided the data into 16 different principal components (there were 16 variables analyzed). Each component represented a new axis in space, each potentially explaining the data set with a different combination of environmental variables without designating some as independent and others as dependent. These represent a measure of the explanatory power of each new axis or principal component (Response variables were not included in the initial analysis). Response variables were regressed against individual principal components. Significance levels for this data were set at $p<0.05$.

RESULTS

Site Characteristics

Mean dissolved oxygen (DO) concentrations were highest at the HC-3 study site (5.4 mg l^{-1}) and the lowest at FC-17 (3.5 mg l^{-1}). Hypoxic events (<2mg l^{-1} of DO) occurred at the FC-17 study site in September (2001), July (2002) and August (2002) and at PC-BDUS in October (2001)). Mean chlorophyll *a* was highest at the HC-SBPGR study site (12.3 μg l^{-1}) and lowest at the HC-3 study site (2.5 μg l^{-1}). However, it should be noted that algal blooms (>25 μg l^{-1} of chlorophyll *a*) occurred at the HC-SBPGR site in June and July of 2002. Mean salinity levels were highest at PC-BDDS (32.1 ppt) and lowest at FC-17 (9.0 ppt). In general flow velocities were greatest at the HC-SBPGR study site and lowest at PC-BDDS with means of 0.12 m s^{-1} and 0.01 m s^{-1} respectively. Nitrate concentrations at the FC-17 study site are high compared to other sites (mean=64.4 μg l^{-1}). The lowest concentrations were at HC-3 (mean=4.1 mg l^{-1}). Ammonium and orthophosphate concentrations were highest at the PC-BDUS study site, mean concentrations were 58.0 μg l^{-1} for ammonium and 17.6 μg l^{-1} for orthophosphate. The lowest concentrations of ammonia occurred at HC-SBPGR (22.5 μg l^{-1}). (It should be noted that the Tidal Creeks Monitoring Program does not run ammonium analysis at the HC-3 study sites, thus these data were not available for this research). The lowest concentration of orthophosphate occurred at HC-3 (6.9 μg l^{-1}). Average N: P ratios were highest at FC-17 (17.3) and lowest at PC-BDDS (7.2). All mean, standard deviation and range data for water quality parameters measured are shown in Table 1.

Table 1. Water quality parameters measured at all study sites, as mean (standard deviation) / (range), July 2001-August 2002. (n=14)

Parameter	FC-17	HC-3	HC-SBPGR	PC-BDDS	PC-BDUS
Temp.	19.5 (4.1) (13.0-26.4)	21.5 (6.5) (10.0-29.6)	21.0 (6.9) (9.4-30.2)	21.2 (6.8) (8.8-29.3)	21.0 (5.5) (12.5-28.5)
DO (mg l^{-1})	3.5 (1.7) (1.0-6.8)	5.4 (2.1) (3.4-9.1)	5.3 (2.3) (2.2-10.3)	4.8 (1.9) (2.4-7.8)	4.2 (1.9) (0.7-7.6)
Salinity (ppt)	9.0 (7.1) (0.7-22.9)	25.5 (6.4) (10.5-34.3)	15.5 (6.2) (3.2-26.2)	32.1 (2.4) (27.2-35.4)	20.9 (9.4) (0.6-31.6)
pH	7.1 (0.4) (6.2-7.6)	7.3 (0.4) (6.5-7.8)	6.9 (0.3) (6.4-7.3)	7.5 (0.5) (6.0-8.0)	7.1 (0.4) (5.9-7.9)
Flow (m s^{-1})	0.1 (0.1) (0.0-0.1)	0.1 (0.0) (0.0-0.1)	0.1 (0.1) (0.1-0.2)	0.0 (0.0) (0.0-0.0)	0.0 (0.0) (0.0-0.0)
Chl. *a* (µg l^{-1})	5.9. (5.0) (0.6-15.9)	2.5 (1.9) (0.5-6.4)	12.3 (24.8) (0.8-51.3)	5.2 (4.2) (0.2-15.2)	8.0 (6.0) (0.7-23.1)
Nitrate (µg l^{-1})	64.4 (58.8) (2.7-173)	4.1 (1.9) (1.8-8.5)	10.4 (10.2) (1.6-35.6)	11.9 (12.9) (2.2-43.9)	10.0 (8.5) (0.0-28.5)
Ammonium (µg l^{-1})	50.7 (78.2) (8.4-288.4)	N/A	22.5 (10.5) (10.8-47.0)	25.9 (11.1) (8.3-42.1)	58.0 (48.3) (16.3-200.0)
Phosphate (µg l^{-1})	14.7 (7.0) (1.9-25.0)	6.9 (2.2) (3.4-11.3)	10.6 (5.2) (4.8-25.8)	11.3 (5.4) (4.9-20.2)	17.6 (5.9) (6.6-32.1)
N:P	17.3 (13.7) (5.3-50.9)	N/A	7.6 (34.4) (2.2-14.9)	7.2 (2.2) (4.2-10.8)	8.8 (5.0) (3.6-21.3)
TSS (mg l^{-1})	21.7 (17.3) (3.3-63.4)	15.6 (9.0) (4.6-32.1)	16.7 (13.2) (1.1-52.7)	21.8 (10.2) (7.0-43.0)	30.5 (21.0) (4.3-80.5)
OSS (mg l^{-1})	2.7 (1.5) (1.2-7.1)	2.0 (0.6) (1.2-3.0)	2.4 (1.0) (0.3-3.9)	2.2 (0.52) (1.3-3.3)	3.4 (2.4) (1.7-10.8)

Suspended and bottom sediment analyses were conducted for each study site. Fluff layer and 10 cm core sample data are shown. Total and organic suspended solids (TSS and OSS) changed seasonally with the highest levels occurring in the summer and spring months. Mean organic suspended solids comprised 15 percent of the total suspended solids measured in these tidal creeks. TSS and OSS concentrations were highest at PC-BDUS, where mean TSS and OSS were 30.5 mg l^{-1} and 3.4 mg l^{-1}, respectively. Suspended solids were the lowest at HC-3 with means of 15.6 mg l^{-1} for TSS and 2.0 mg l^{-1} for OSS (Table 1).

Modal grain size of bottom sediments changed very little over the sampling period (Fig. 5). Bottom sediment grain sizes at the FC-17, HC-3 and PC-BDUS study sites were coarser that grain sizes at other sites. The coarsest material, fine sands, occurred at PC-BDUS (175.5 μm). The finest grain sizes, silty mud, occurred at HC-SBPGR (10.0 μm). Organic percentages varied little over the sampling period (Fig. 6). Mean organic content of bottom sediments was highest at HC-SBPGR (18.3%) and lowest at PC-BDUS (2.8%). Mean ratios of total: soluble carbohydrate were low for sand-dominated sediments and high for silty mud sediments (Fig. 7). The highest ratio occurred in bottom sediments at site HC-SBPGR (101.2) and the lowest ratio was found at HC-3 (25.2).

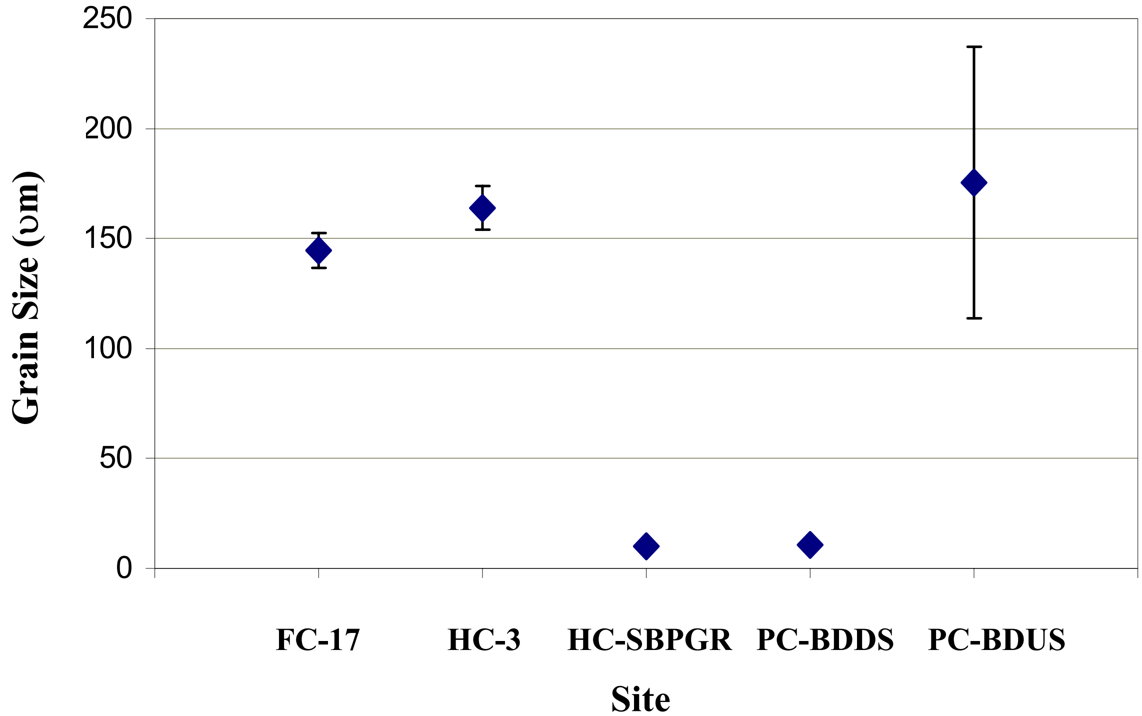

Figure 5. Seasonal modal grain size (10 cm core) for all study sites over the sampling period July 2001-Aug. 2002.

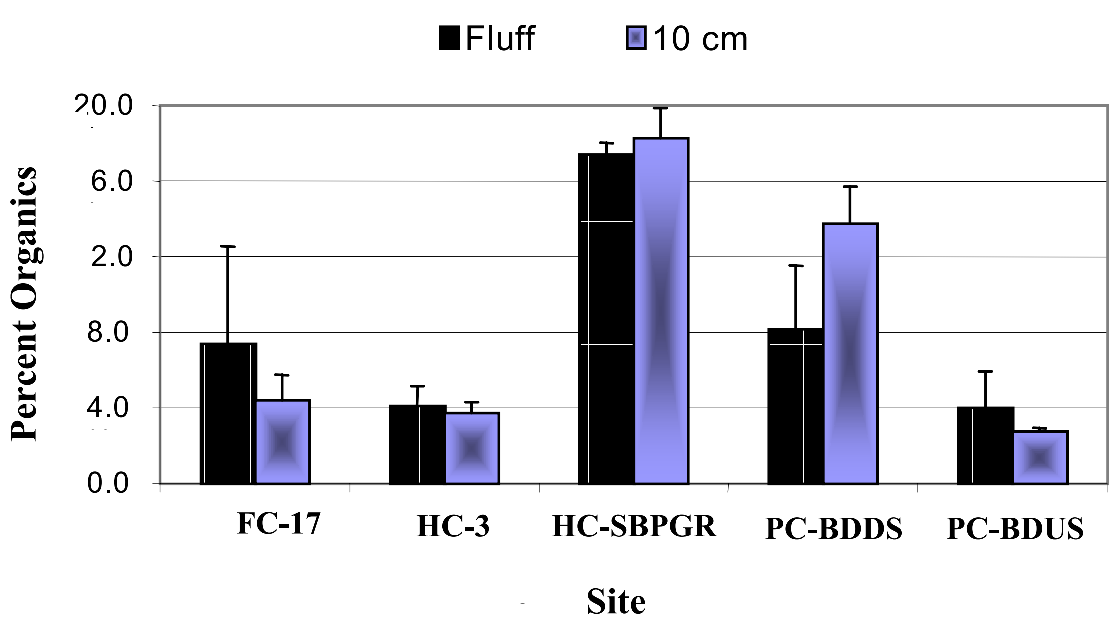

Figure 6. Seasonal percent of organic material in bottom sediments at all study sites

Table 2. Mean total: soluble carbohydrate ratios of bottom sediment for all study sites. (Tot: Sol=Ratio of total to soluble carbohydrates).

Date	Site	Total Carbohydrates (μg/g)	Solyble Carbohydrates (μg/g)	Tot.: Sol.
Jul-01	FC-17	10725.0	288.1	37.2
Oct-01	FC-17	11785.9	291.8	40.4
Mar-02	FC-17	14316.3	449.6	29.0
Jun-02	FC-17	26110.7	359.9	72.6
Jul-01	HC-3	7031.1	354.3	19.8
Oct-01	HC-3	2146.5	187.7	11.4
Mar-02	HC-3	10016.3	199.5	50.2
Jun-02	HC-3	7848.2	406.5	19.3
Jul-01	HCSBPGR	28837.5	221.9	130.0
Oct-01	HC-SBPGR	38072.2	392.0	97.1
Mar-02	HC-SBPGR	31384.1	791.4	39.7
Jun-02	HC-SBPGR	47012.9	415.9	113.0
Jul-01	PC-BDDS	2713.9	209.3	13.0
Oct-01	PC-BDDS	41090.8	391.7	104.9
Mar-02	PC-BDDS	29517.3	191.5	154.2
Jun-02	PC-BDDS	23741.1	483.9	49.1
Jul-01	PC-BDUS	25313.5	493.8	51.3
Oct-01	PC-BDUS	4106.2	146.1	28.1
Mar-02	PC-BDUS	14316.3	401.8	35.6
Jun-02	PC-BDUS	8210.9	231.3	35.5

Oxygen Demand

Biochemical Oxygen Demand

Rates of five-day biochemical oxygen demand (BOD_5) for all sites over the entire sampling period varied from 0.0 to 7.6 mg l^{-1} (Fig. 7). Yearly mean BOD_5 rates did not significantly differ between study sites (p=0.2842). A maximum rate of 7.6 mg l^{-1} occurred at the south branch study site in Hewletts Creek (HC- SBPGR) in June (2002) and the minimum rate of 0.0 mg l^{-1} was found at the mid-creek station (HC-3) in the lower part of Hewletts Creek in October (2001). Mean rates ranged from 2.0 to 3.0 mg 1^{-1}.

Sediment Oxygen Demand

Rates of sediment oxygen uptake for all study sites ranged from –1.5 to 6.3 g O_2 m^{-2} d^{-1} (Fig. 8). A Kruskal-Wallis nonparametric test was used to determine that mean SOD rates were significantly different among study sites (p=0.0070). The results of a Wilcoxon 2-sample test indicate that yearly mean SOD rates at PC-BDDS were significantly greater than rates at FC-17 (p=0.0013), but yearly mean SOD rates at other sites did not significantly differ. The maximum rate of 6.3 g O_2 m^{-2} d^{-1} occurred at the upstream tributary site in Pages Creek (PC-BDDS) in November (2001) and the minimum rate of –1.5 g O_2 m^{-2} d^{-1} occurred in the downstream tributary site of Pages Creek (PC-BDUS) in July (2002). Mean rates ranged from 0.0 to 1.9 g O_2 m^{-2} d^{-1}. Mean rates were the highest at an upper tributary site in Pages Creek (PC-BDDS) and the lowest at FC-17, an upper tributary site in Futch Creek.

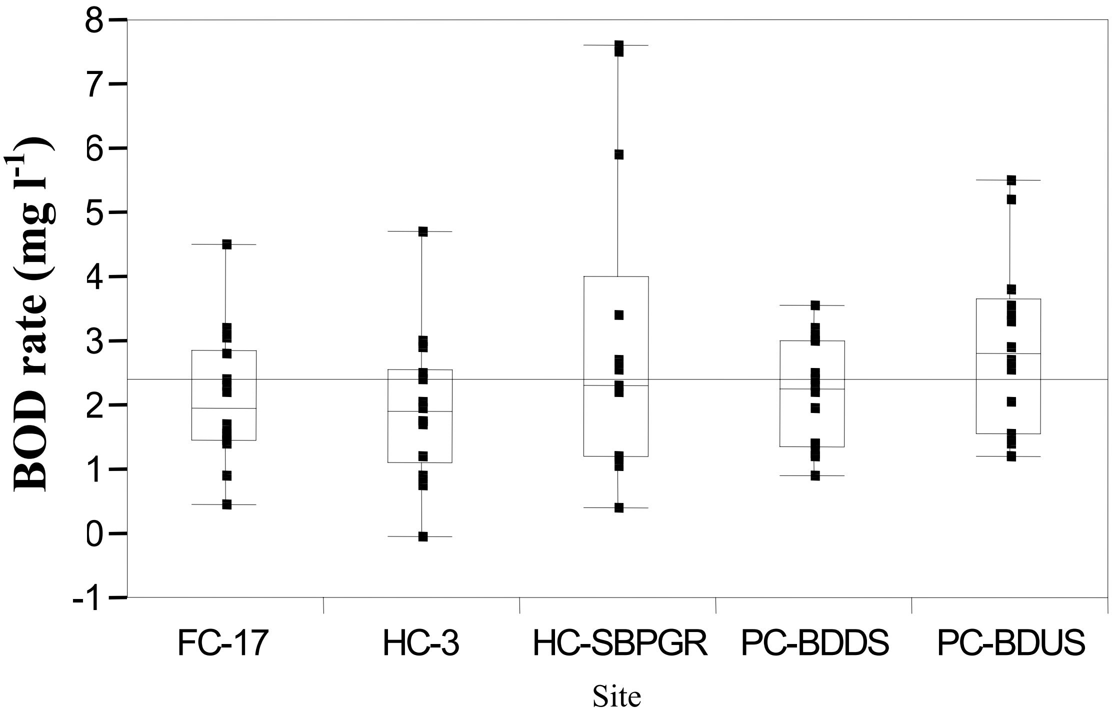

Figure 8. Box plots of mean, range and confidence intervals for BOD_5 data by site for the sampling period July 2001-Aug. 2002.

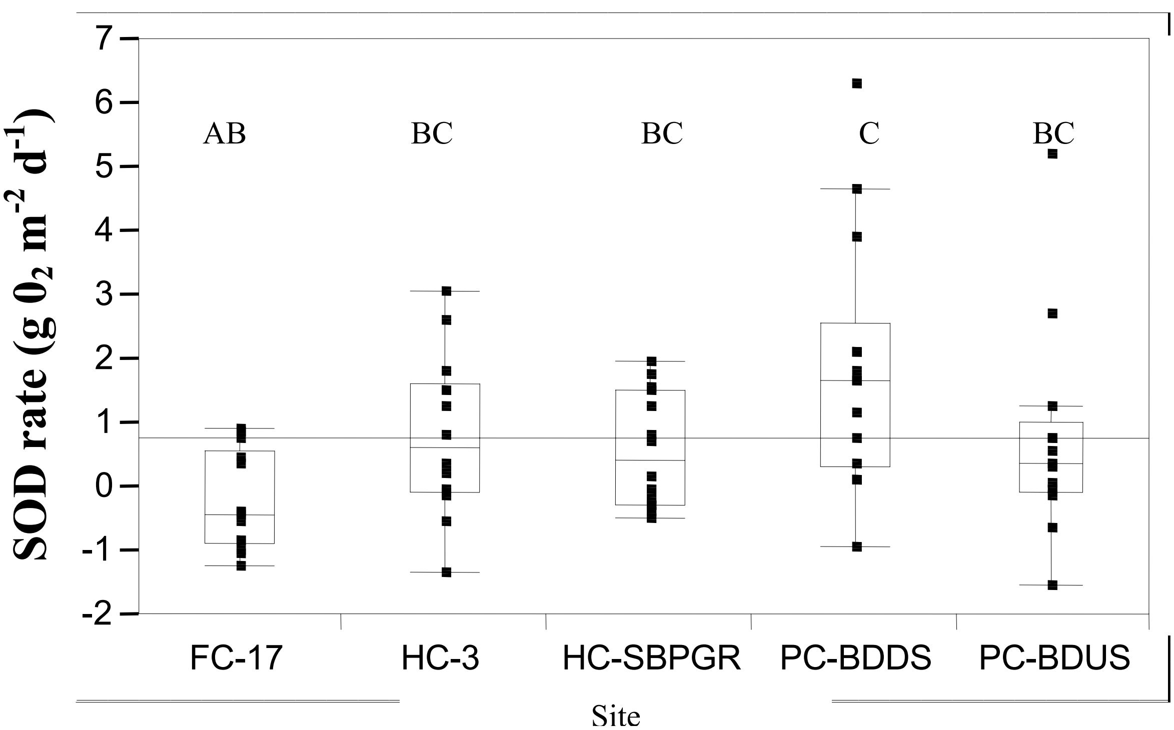

Figure 8. Box plots of mean, range and confidence intervals for SOD data by site for the sampling period July 2001-Aug. 2002. Different letters indicate significant differences among stations.

Seasonal Oxygen Demand

Mean SOD rates were greater than mean BOD_5 rates at all study sites when converted to common units ($g\ O_2\ m^{-2}\ d^{-1}$) (Table 3). A large amount of temporal variation existed in SOD rates while BOD_5 rates appeared more constant over time (Fig. 9). SOD, therefore, appeared to be a larger and more variable sink for oxygen than BOD.

BOD_5 rates were highest in summer and spring and lowest in fall and winter, whereas SOD rates were highest in the winter and spring months and lowest in the fall and summer months (Fig. 10). As temperatures increased so did the water-column demand for oxygen, probably due to an increase in respiration in the biotic community. The solubility of oxygen decreased as temperature increased so increased respiration was coupled with a decrease of the capacity of water to dissolve oxygen in the summer months. Although SOD appeared to be more dominant in the winter months, a decrease in BOD occurred when water temperatures cooled and biological activity was greatly reduced.

Correlation Analyses for Individual Study Sites

Significant correlations between environmental variables for each of the FC-17, HC-3, HC-SBPGR, PC-BDDS and PC-BDUS study sites are shown in Tables 4-8, respectively. Numerous variables co-varied in this data set, thus it was inappropriate to try to predict specific relationships using pair-wise correlations. A principal components analysis was conducted in SAS (1998) to resolve correlation patterns in the data set.

Table 3. BOD_5 and SOD mean and standard deviation data for each study site. (Rates are in the same units for comparison).

	BOD_5 g O_2 m^{-2} d^{-1}	SOD g O_2 m^{-2} d^{-1}
FC-17	0.14 ± 0.10	0.23 ± 0.76
HC-3	0.15 ± 0.07	0.81 ± 1.24
HC-SBPGR	0.26 ± 0.23	0.59 ± 0.89
PC-BDDS	0.19 ± 0.06	1.85 ± 1.96
PC-BDUS	0.27 ± 0.15	0.76 ± 1.67

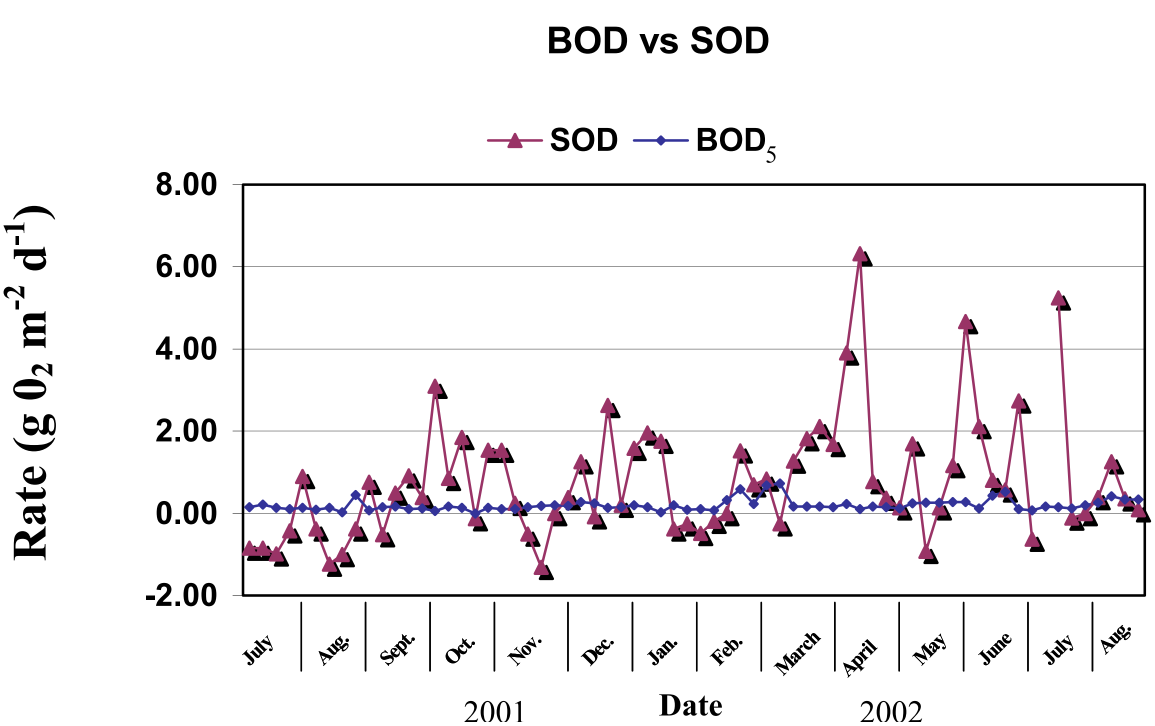

Figure 9. Oxygen demand rates on an areal basis for the 2001-2002 sampling season.

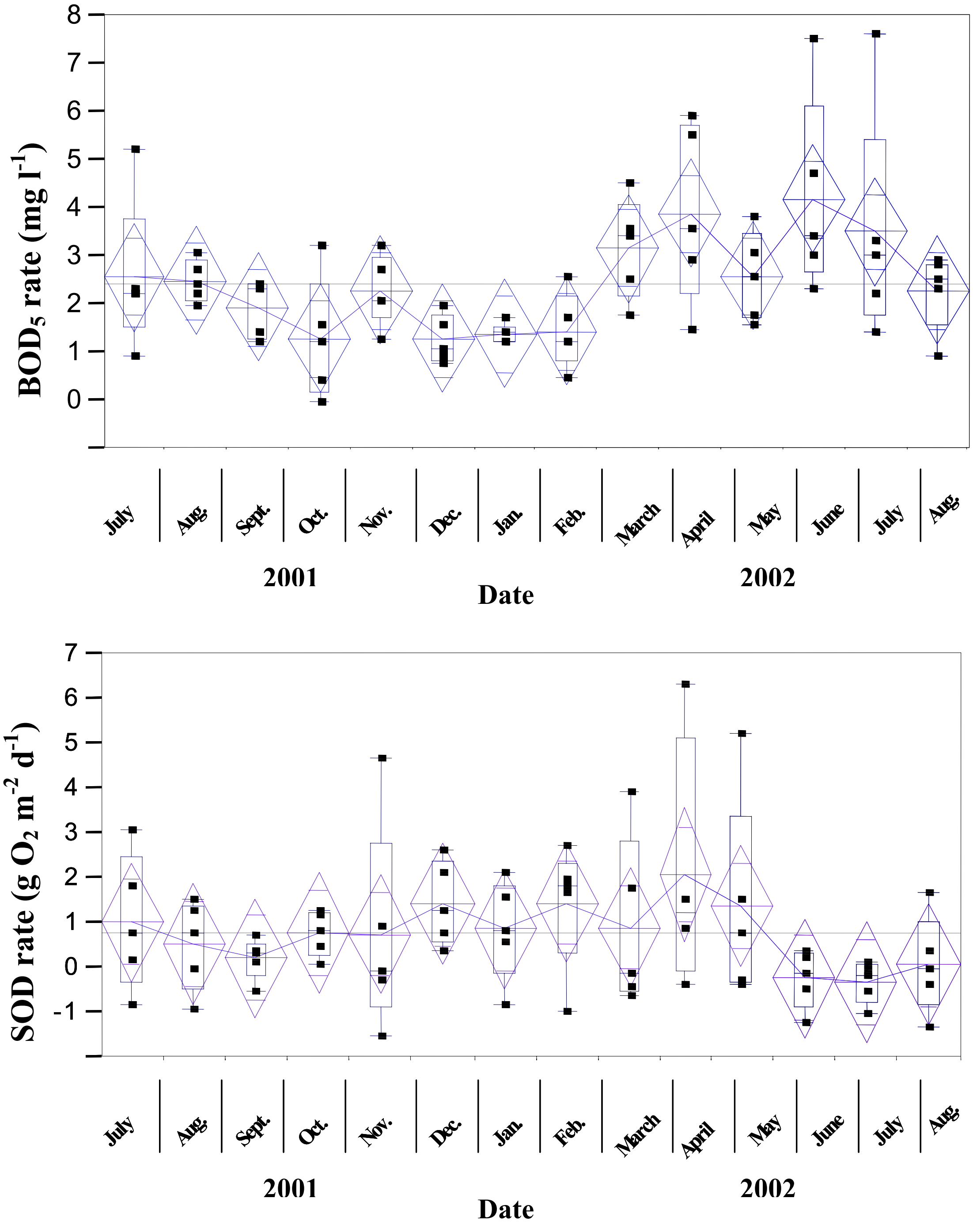

Figure 10. BOD_5 and SOD rates for all sites combined over time.

Table 4. Correlation matrix for study site FC-17 (Log transformations are indicated by L). Shaded areas represent significance with $p<0.05$.

	Year	Depth	Temp	LDO	pH	Salinity	SOD	$LBOD_5$	LChl. a	LNH4	LNIT	LPO_4	N:P	LTSS	OSS	Org C	Tot:Sol
Year	1.00																
Depth	0.02	1.00															
Temp	-0.06	0.06	1.00														
LDO	0.01	0.23	-0.63	1.00													
pH	0.12	-0.11	-0.36	0.22	1.00												
Salinity	0.54	0.34	0.44	-0.45	0.12	1.00											
SOD	0.25	-0.45	0.46	-0.49	0.30	*0.55	1.00										
$LBOD_5$	-0.14	0.21	0.24	-0.39	-0.44	0.13	0.15	1.00									
LChl. a	0.08	0.17	*0.72	*-0.59	-0.52	0.24	0.31	*0.62	1.00								
LNH4	0.20	0.21	*0.65	-0.09	0.03	0.35	0.42	0.48	0.48	1.00							
LNIT	0.24	0.35	0.01	0.40	-0.11	-0.09	-0.32	-0.39	-0.06	0.47	1.00						
LPO_4	-0.08	-0.04	0.28	0.37	-0.02	-0.21	-0.18	-0.50	-0.18	0.46	*0.71	1.00					
N:P	0.48	*0.61	0.05	-0.05	-0.07	0.33	0.11	0.33	0.46	0.48	0.41	-0.24	1.00				
LTSS	0.02	0.16	*0.68	*0.60	-0.47	0.35	0.32	*0.60	*0.56	0.52	-0.02	0.07	0.24	1.00			
OSS	-0.37	-0.04	*0.66	0.07	*-0.73	-0.26	-0.11	0.57	0.41	0.19	-0.20	0.09	-0.17	*0.70	1.00		
Org C	-0.24	-0.48	0.56	*-0.99	0.82	-0.03	*0.88	0.31	0.78	0.74	-0.31	-0.11	0.44	-0.05	0.11	1.00	
Tot:Sol	0.33	-0.48	0.39	-0.66	0.25	-0.18	0.73	0.66	0.60	0.91	0.61	0.74	*0.97	0.33	0.31	0.56	1.00
D	-0.19	0.43	*0.72	-0.33	-0.45	0.35	0.05	0.04	0.40	*0.55	0.43	0.43	0.22	*0.53	0.20	-0.10	-0.09
D 24	0.28	0.33	0.32	-0.37	-0.26	0.54	0.00	0.16	0.38	0.35	0.30	-0.15	*0.64	0.25	-0.18	-0.21	-0.25
D 48	0.21	0.25	0.23	-0.09	0.16	*0.59	0.29	-0.24	0.02	0.30	0.18	0.07	0.09	-0.15	-0.34	0.13	-0.59
D 72	0.20	0.13	0.40	-0.08	-0.21	0.35	0.22	-0.14	0.09	0.47	0.50	0.41	0.23	0.31	0.06	-0.71	-0.03
Rain 24	-0.12	0.38	*0.71	-0.35	-0.38	0.40	0.17	0.00	0.37	0.54	0.44	0.42	0.24	0.49	0.12	-0.10	-0.09
Rain 48	0.01	0.44	*0.62	-0.29	-0.25	*0.54	0.13	-0.03	0.35	0.51	0.41	0.27	0.30	0.34	-0.04	-0.02	-0.54
Rain 72	0.10	0.32	*0.56	-0.21	-0.24	0.47	0.18	-0.09	0.25	0.51	0.48	0.35	0.27	0.35	0.00	-0.55	-0.70

Note: For all correlation matrices: LDO=log dissolved oxygen, $LBOD_5$=Log BOD_5, LChl. *a*=Log chlorophyll *a*, LNH_4=Log Ammonium, LNIT=Log Nitrate, LPO_4=Log Orthophosphate, N:P=ratio of nitrogen to orthophosphate, LTSS=Log of total suspended solids, OSS= Organic suspended solids, Org C= Percent Organic Content, Tot:Sol=Ratio of total to Soluble Carbohydrates, D, D24, D48 and D72=Rainfall on day of sampling, 24 hours prior, 48 hours prior and 72 hours prior respectively, and Rain 24, Rain 48 and Rain 72= Cumulative rainfall 24, 48 and 72 hours prior to sampling.

Table 5. Correlation matrix for study site HC-3 (Log transformations are indicated by L). Shaded areas represent significance with $p<0.05$.

	Year	Depth	Temp	LDO	pH	Salinity	SOD	$LBOD_5$	LChl. *a*	LNIT	LPO_4	LTSS	OSS	Org C	Tot:Sol
Year	1.00														
Depth	0.42	1.00													
Temp	-0.15	-0.30	1.00												
LDO	0.02	0.16	*-0.94	1.00											
pH	-0.36	-0.44	-0.38	0.47	1.00										
Salinity	-0.05	0.19	-0.23	0.23	0.36	1.00									
SOD	-0.47	*-0.68	0.45	-0.28	0.37	0.05	1.00								
$LBOD_5$	0.51	-0.20	0.22	-0.29	-0.28	-0.44	0.05	1.00							
LChl. *a*	-0.06	-0.14	*0.88	*-0.91	-0.51	-0.37	0.48	0.23	1.00						
LNIT	-0.40	-0.26	-0.13	0.05	0.44	0.01	0.36	-0.27	0.00	1.00					
LPO_4	-0.24	-0.50	0.22	-0.15	0.27	-0.34	0.25	0.09	0.13	0.32	1.00				
LTSS	-0.16	-0.60	*0.82	*-0.74	-0.12	-0.40	*0.67	0.51	*0.79	-0.01	0.32	1.00			
OSS	-0.20	-0.36	0.73	*-0.73	-0.48	-0.59	0.28	0.36	*0.73	-0.98	0.27	*0.82	1.00		
Org C	-0.52	-0.18	0.67	-0.14	-0.39	-0.87	0.85	-0.11	0.65	0.89	0.57	0.44	0.00	1.00	
Tot:Sol	0.65	0.81	-0.83	0.96	-0.97	0.36	-0.57	0.32	-0.74	-0.61	-0.91	-0.65	-0.85	-0.20	1.00
D	-0.39	-0.36	-0.10	-0.07	0.40	0.26	0.24	-0.08	-0.14	*0.62	0.15	-0.02	-0.43	-0.42	*0.97
D 24	0.14	-0.35	0.08	-0.17	0.10	-0.70	0.18	0.50	0.25	0.40	0.22	0.40	0.24	-0.26	-0.23
D 48	-0.28	-0.06	0.37	-0.11	0.05	-0.69	*0.56	-0.17	*0.46	0.22	0.16	0.29	0.06	*0.99	-0.26
D 72	0.13	0.15	0.39	-0.40	-0.16	0.03	-0.20	0.26	0.13	-0.33	0.25	0.17	0.25	*0.99	-0.21
Rain 24	-0.18	-0.46	-0.02	-0.15	0.34	-0.25	0.28	0.25	0.05	*0.68	0.24	0.23	-0.11	-0.30	-0.15
Rain 48	-0.32	-0.43	0.20	-0.15	0.32	-0.31	*0.56	0.12	0.30	*0.71	0.29	0.36	-0.04	0.62	-0.33
Rain 72	-0.18	-0.27	0.47	-0.44	0.15	-0.23	0.34	0.30	0.37	0.37	0.45	0.45	0.18	0.63	-0.33

Table 6. Correlation matrix for study site HC-SBPGR (Log transformations are indicated by L). Shaded areas represent significance with p<0.05.

	Year	Depth	Temp	LDO	pH	Salinity	SOD	$LBOD_5$	LChl. a	LNH4	LNIT	LPO_4	N:P	LTSS	OSS	Org C	Tot:Sol
Year	1.00																
Depth	0.05	1.00															
Temp	-0.09	0.00	1.00														
LDO	0.18	-0.09	*-0.86	1.00													
pH	-0.04	0.37	-0.29	0.35	1.00												
Salinity	-0.44	-0.16	0.07	0.26	0.51	1.00											
SOD	-0.23	-0.32	0.50	*-0.78	-0.42	0.14	1.00										
$LBOD_5$	0.47	-0.01	0.50	-0.28	0.09	-0.34	-0.02	1.00									
LChl. a	0.16	-0.03	*0.78	*-0.60	-0.11	-0.29	0.45	*0.72	1.00								
LNH4	0.10	0.13	-0.08	-0.03	0.04	0.44	-0.12	-0.31	-0.39	1.00							
LNIT	*-0.56	0.21	-0.05	-0.02	0.25	0.13	-0.02	*-0.64	-0.27	0.29	1.00						
LPO_4	0.00	-0.10	-0.03	-0.09	0.25	-0.42	0.15	-0.01	0.04	0.18	0.36	1.00					
N:P	-0.12	0.12	0.15	-0.05	-0.16	0.72	-0.12	-0.36	-0.22	0.60	0.31	-0.49	1.00				
LTSS	0.29	0.11	*0.78	*-0.65	-0.30	-0.34	0.40	*0.53	*0.83	-0.23	-0.11	-0.07	0.00	1.00			
OSS	0.33	0.21	*0.60	-0.31	-0.17	-0.35	-0.01	0.69	0.68	-0.46	-0.42	-0.07	-0.27	0.58	1.00		
Org C	0.62	0.83	-0.67	-0.83	0.43	*-0.99	0.41	0.20	0.09	-0.82	-0.86	0.01	-0.91	-0.37	0.16	1.00	
Tot:Sol	-0.51	-0.74	*0.99	*-0.99	-0.19	-0.76	0.12	0.05	0.43	0.77	0.57	0.78	-0.90	0.66	0.25	-0.61	1.00
D	-0.39	-0.04	-0.09	-0.14	0.23	0.33	0.17	-0.21	-0.22	0.09	0.21	-0.22	0.24	-0.09	*-0.64	0.40	-0.85
D 24	0.14	0.08	0.10	-0.21	0.42	*-0.57	0.21	0.39	0.39	-0.20	-0.07	0.52	-0.49	0.23	0.05	0.45	0.28
D 48	-0.28	-0.12	0.40	-0.16	0.43	*-0.63	-0.11	0.02	0.25	-0.11	*0.53	0.15	0.24	0.27	0.20	*-0.96	0.77
D 72	0.13	0.36	0.39	-0.18	-0.13	-0.20	-0.18	0.44	0.39	0.00	-0.20	-0.15	-0.07	0.48	0.46	*-0.99	0.69
Rain 24	-0.18	0.02	0.00	-0.23	0.42	-0.12	0.25	0.09	0.02	-0.06	0.10	0.16	-0.13	0.07	-0.41	0.48	0.22
Rain 48	-0.32	-0.05	0.23	-0.23	0.43	-0.18	0.15	0.09	0.21	-0.12	0.39	0.22	0.02	0.21	-0.24	-0.45	0.81
Rain 72	-0.18	0.23	0.50	-0.33	0.27	-0.31	0.00	0.15	0.49	-0.10	0.19	0.08	-0.03	*0.56	0.14	-0.46	0.82

Table 7. Correlation matrix for study site PC-BDDS (Log transformations are indicated by L). Shaded areas represent significance with $p<0.05$.

	Year	Depth	Temp	LDO	pH	Salinity	SOD	$LBOD_5$	LChl.a	LNH4	LNIT	LPO_4	N:P	LTSS	OSS	Org C	Tot:Sol
Year	1.00																
Depth	0.13	1.00															
Temp	-0.18	0.13	1.00														
LDO	-0.16	-0.02	*-0.89	1.00													
pH	0.11	*-0.65	-0.56	0.41	1.00												
Salinity	-0.23	-0.37	-0.13	0.26	0.51	1.00											
SOD	-0.42	0.11	0.22	0.03	0.08	-0.05	1.00										
$LBOD_5$	0.04	-0.34	0.25	-0.31	-0.30	-0.20	-0.20	1.00									
LChl. a	-0.17	0.00	*0.86	*-0.728	-0.37	-0.02	0.37	0.39	1.00								
LNH4	-0.47	0.21	-0.37	0.53	-0.07	-0.01	0.25	-0.11	-0.17	1.00							
LNIT	*-0.76	0.10	0.28	0.05	-0.46	-0.06	0.10	0.23	0.28	0.50	1.00						
LPO_4	*-0.65	-0.12	-0.15	0.38	-0.12	0.18	0.10	0.12	-0.10	*0.81	*0.58	1.00					
N:P	-0.10	0.63	0.16	-0.02	-0.25	-0.23	0.27	-0.20	0.27	0.31	0.45	-0.18	1.00				
LTSS	-0.13	0.13	*0.52	-0.30	-0.52	0.16	0.02	0.46	0.45	0.03	0.12	0.26	-0.21	1.00			
OSS	-0.46	0.11	*0.634	-0.41	*-0.74	-0.04	0.06	*0.66	*0.59	0.07	*0.58	0.38	-0.01	*0.78	1.00		
Org C	-0.29	-0.48	-0.84	0.77	0.94	0.07	0.76	0.72	0.22	0.00	0.21	0.40	0.95	-0.80	-0.09	1.00	
Tot:Sol	0.34	0.01	-0.81	0.58	0.28	1.00	-0.19	0.78	-0.70	0.98	-0.71	-0.51	-0.39	0.22	0.69	0.41	1.00
D	0.32	0.16	0.42	*-0.58	-0.12	*-0.58	0.33	0.17	0.38	-0.26	-0.17	-0.44	0.18	-0.09	-0.05	-0.11	-0.35
D 24	-0.07	0.27	0.52	*-0.68	-0.44	*-0.62	0.31	-0.03	0.31	-0.36	0.02	-0.20	0.13	-0.04	0.20	-0.10	-0.34
D 48	0.22	0.30	0.32	-0.44	-0.17	*-0.63	0.42	0.05	0.21	-0.24	-0.17	-0.36	0.31	-0.07	-0.06	-0.09	-0.30
D 72	-0.01	-0.11	0.45	-0.37	-0.12	-0.30	0.09	0.23	0.20	-0.14	0.09	-0.26	-0.05	0.21	0.20	0.77	-0.27
Rain 24	0.14	0.23	0.51	*-0.69	-0.29	*-0.65	0.35	0.08	0.38	-0.54	-0.08	-0.35	0.17	-0.07	0.06	-0.11	-0.35
Rain 48	0.16	0.24	0.49	*-0.66	-0.28	*-0.66	0.36	0.78	0.36	-0.33	-0.10	-0.36	0.19	-0.07	0.04	-0.11	-0.35
Rain 72	0.13	0.17	*0.562	*-0.645	-0.26	*-0.62	0.34	0.14	0.38	-0.39	-0.05	-0.39	0.15	-0.01	0.10	-0.89	-0.64

Table 8. Correlation matrix for study site PC-BDUS (Log transformations are indicated by L). Shaded areas represent significance with $p<0.05$.

	Year	Depth	Temp	LDO	pH	Salinity	SOD	$LBOD_5$	LChl.a	LNH4	LNIT	LPO_4	N:P	LTSS	OSS	Org C	Tot:Sol
Year	1.00																
Depth	0.04	1.00															
Temp	-0.11	0.43	1.00														
LDO	0.11	-0.16	-0.45	1.00													
pH	0.22	*-0.71	*-0.60	0.38	1.00												
Salinity	0.51	0.53	0.02	0.43	-0.08	1.00											
SOD	-0.48	-0.18	-0.25	0.33	0.04	-0.01	1.00										
$LBOD_5$	0.43	0.23	0.47	-0.04	-0.33	0.49	-0.25	1.00									
LChl. a	0.11	0.31	*0.85	-0.44	-0.34	-0.09	-0.43	0.35	1.00								
LNH4	-0.16	-0.12	-0.27	0.09	-0.06	-0.09	0.01	-0.08	-0.19	1.00							
LNIT	*-0.60	0.20	0.45	-0.18	-0.33	-0.24	0.15	-0.14	0.18	0.23	1.00						
LPO_4	-0.09	-0.33	-0.46	0.26	0.01	-0.13	0.17	0.11	-0.49	*0.68	-0.29	1.00					
N:P	0.07	0.06	0.15	-0.10	-0.20	0.01	-0.14	0.14	0.20	*0.86	0.26	0.25	1.00				
LTSS	0.28	0.46	0.46	0.21	-0.25	*0.75	0.03	*0.60	0.37	-0.06	-0.19	-0.06	0.21	1.00			
OSS	0.19	0.13	0.52	-0.07	-0.02	0.25	-0.11	0.33	*0.57	0.09	0.32	-0.17	0.36	0.51	1.00		
Org C	-0.19	-0.23	-0.12	-0.39	0.97	-0.31	-0.72	-0.64	0.33	*-0.99	0.28	-0.78	-0.92	-0.43	0.42	1.00	
Tot:Sol	-0.24	0.39	0.85	0.99	-0.29	0.98	0.90	0.86	-0.08	0.54	0.71	0.26	0.81	0.64	0.10	-0.49	1.00
D	0.32	0.27	0.45	-0.23	0.06	0.14	-0.42	0.17	*0.55	-0.37	0.11	*-0.78	0.08	0.19	0.44	0.55	-0.13
D 24	-0.07	0.25	0.46	-0.18	-0.19	-0.29	-0.15	-0.11	0.52	-0.46	0.23	*-0.68	-0.11	-0.09	0.07	0.56	-0.14
D 48	-0.22	0.38	0.35	-0.28	-0.08	0.10	-0.42	0.13	0.38	-0.47	-0.03	*-0.75	-0.09	0.07	-0.05	0.56	-0.13
D 72	-0.01	0.11	0.51	-0.27	-0.15	0.09	-0.27	0.52	0.29	0.09	0.28	-0.19	0.49	0.27	0.05	-0.80	0.88
Rain 24	0.14	0.29	0.50	-0.23	-0.06	-0.07	-0.32	0.04	*0.58	-0.45	0.17	*-0.80	-0.01	0.06	0.30	0.55	-0.13
Rain 48	0.16	0.30	0.48	-0.24	-0.07	-0.04	-0.33	0.05	*0.57	-0.46	0.16	*-0.80	-0.02	0.07	0.25	0.55	-0.13
Rain 72	0.13	0.29	*0.57	-0.26	-0.09	-0.02	-0.37	0.21	*0.57	-0.37	0.22	*-0.75	0.09	0.14	0.23	-0.23	0.75

Correlation Analysis for All Sites Combined

For the entire 14-month sampling period there were numerous significant correlations (Table 9). For example, DO and chlorophyll *a* were correlated negatively and positively, respectively, with temperature. Rainfall data, which was below average during the study year, showed some correlations with DO, chlorophyll *a*, and BOD_5 measurements (Fig. 11). Dissolved oxygen was negatively correlated with rainfall over the 24-48 hours previous to sampling. Chlorophyll *a* positively correlated with rainfall 24 hours before sampling and BOD_5 was correlated with rainfall 72 hours prior to sampling. BOD_5 was positively correlated with Chlorophyll *a*; this relationship is illustrated in Figure 12.

There was a significant positive relationship between SOD rates and percent of organic content in bottom sediments (Fig. 13). There was no significant correlation between SOD and the amount of soluble (bioavailable) carbohydrates in the bottom sediment for all study sites combined, however, the organic content of bottom sediments was positively correlated with total: soluble carbohydrate ratios.

Numerous significant correlations among explanatory and response variables exist in this data set. May variables co-vary making it inappropriate to try to understand specific relationships using simple pair-wise correlations. Thus, a principal component analysis was conducted to reduce the number of variables in the data.

Table 9. Correlation matrix for all study sites (Log transformations are indicated by L). Shaded areas represent significance with $p<0.05$.

	Year	Depth	Temp	LDO	pH	Salinity	SOD	$LBOD_5$	LChl.a	LNH4	LNIT	LPO_4	N:P	LTSS	OSS	Org C	Tot:Sol
Year	1.00																
Depth	0.09	1.00															
Temp	-0.12	0.13	1.00														
LDO	0.04	0.04	*-0.65	1.00													
pH	0.02	-0.23	*-0.39	*0.33	1.00												
Salinity	0.09	*0.44	0.08	0.20	*0.31	1.00											
SOD	*-0.28	0.02	0.23	-0.02	0.21	*0.36	1.00										
$LBOD_5$	*0.28	0.05	*0.34	-0.20	*-0.26	-0.06	-0.06	1.00									
LChl. a	0.03	0.10	*0.74	*-0.57	*-0.38	-0.15	0.14	*0.53	1.00								
LNH4	-0.03	0.11	0.01	-0.03	-0.02	0.08	0.09	-0.04	0.04	1.00							
LNIT	*-0.27	-0.01	0.02	-0.21	-0.17	*-0.42	-0.16	-0.21	0.06	*0.30	1.00						
LPO_4	-0.16	-0.07	-0.05	-0.09	-0.04	*-0.26	0.00	0.04	0.05	*0.56	*0.44	1.00					
N:P	0.20	0.04	-0.01	-0.18	-0.12	-0.16	-0.17	0.00	0.13	*0.44	*0.54	-0.10	1.00				
LTSS	0.08	0.22	*0.59	-0.32	-0.21	0.20	*0.24	*0.52	*0.58	0.21	-0.03	0.18	0.09	1.00			
OSS	-0.01	0.09	*0.40	-0.21	*-0.28	-0.11	-0.08	*0.45	*0.50	0.15	0.08	0.20	0.00	*0.57	1.00		
Org C	-0.02	0.04	0.08	0.30	0.18	0.14	*0.63	0.13	0.37	*-0.54	-0.34	-0.26	*-0.56	0.06	-0.14	1.00	
Tot:Sol	0.10	-0.03	0.00	0.22	0.25	0.25	0.14	0.24	0.24	-0.41	-0.06	-0.04	-0.29	0.17	-0.01	*0.44	1.00
D	-0.01	0.15	*0.27	*-0.29	-0.01	0.10	0.02	0.01	0.23	0.04	0.19	-0.07	0.11	0.15	0.15	-0.12	-0.17
D 24	0.03	*0.23	*0.32	*-0.26	-0.09	-0.07	0.19	0.17	*0.34	-0.10	0.01	-0.05	-0.03	0.14	0.06	0.20	0.05
D 48	-0.05	0.02	*0.30	*-0.27	0.00	-0.12	0.06	-0.09	0.20	-0.01	0.21	-0.04	0.17	0.06	-0.06	0.01	0.05
D 72	0.10	0.09	*0.39	-0.23	-0.16	-0.04	-0.08	*0.24	0.21	0.16	0.16	0.09	0.21	*0.30	0.14	-0.12	-0.05
Rain 24	0.00	0.21	*0.34	*-0.32	-0.05	0.03	0.12	0.08	*0.31	-0.03	0.12	-0.05	0.02	0.16	0.13	0.03	-0.10
Rain 48	-0.01	0.19	*0.39	*-0.33	-0.05	0.00	0.12	0.05	*0.34	-0.03	0.18	-0.07	0.10	0.16	0.09	0.03	-0.05
Rain 72	0.04	0.20	*0.48	*-0.35	-0.10	-0.02	0.06	0.15	*0.36	0.04	0.21	-0.02	0.16	*0.26	0.13	-0.06	-0.09

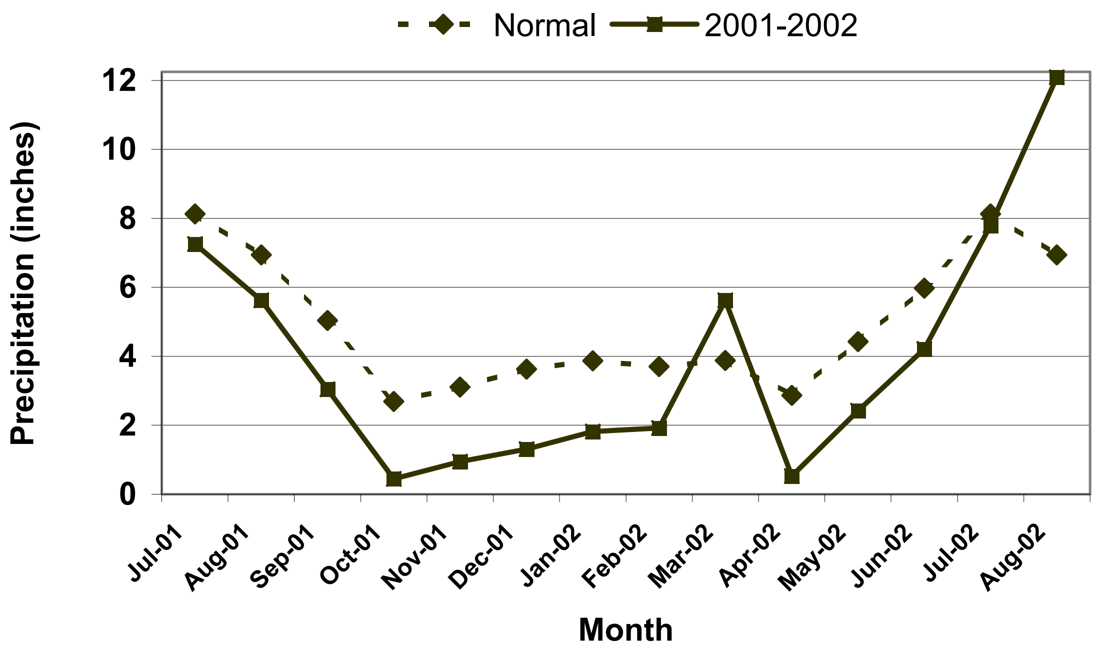

Figure 11. Mean monthly rainfall over the sampling period compared to long-term averages (NOAA).

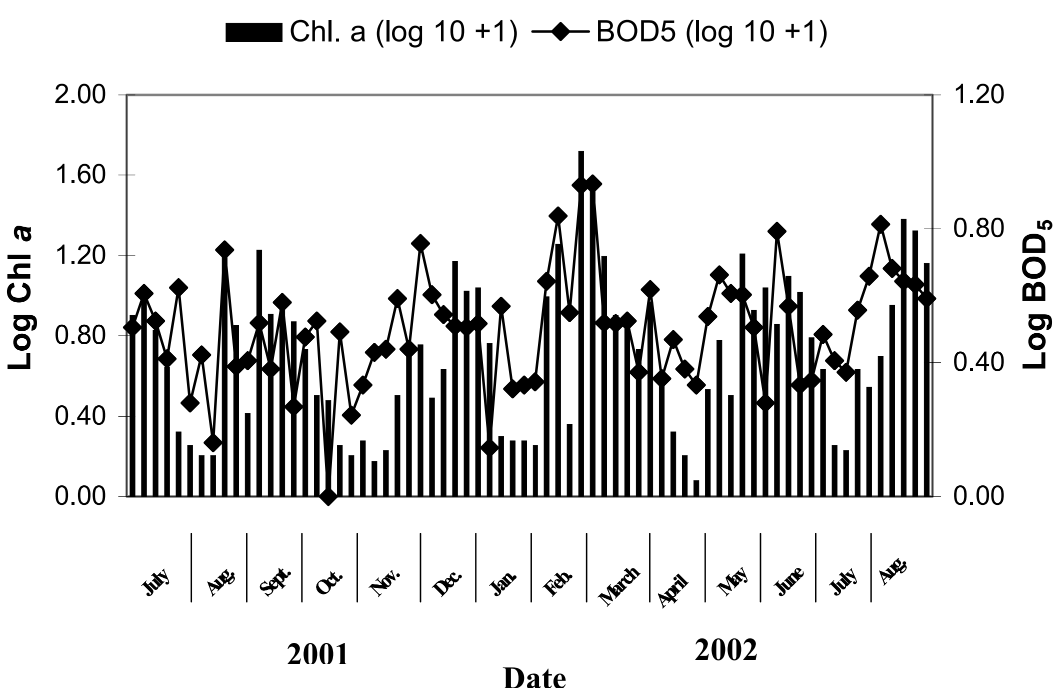

Figure 12. Mean Log BOD_5 and Log Chlorophyll *a* values for all sites vs. Time.

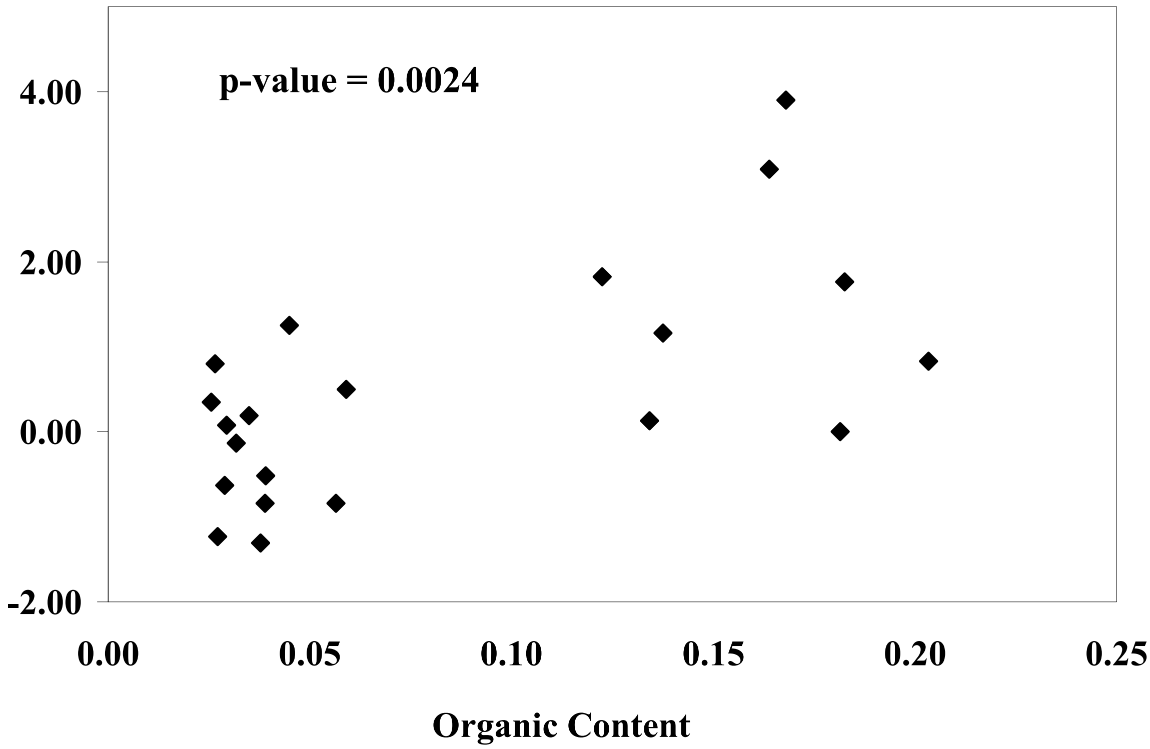

Figure 13. Scatter plot of percent organic content of bottom sediments and SOD rates for all sites combined.

Principal Components Analysis (PCA)

A PCA analysis of the 16 environmental variables analyzed was conducted using SAS statistical software. Principal component 1 alone explained 30 percent of the variability in the data set while principal components 1-4 together explained 74 percent of the data (Table 10). Adding a fifth principal component only increased this cumulative percentage by 6 percent, thus the PCA analysis focused on principal components 1-4. However, since significant regressions only occurred between response variables and principal components 1-3 principal component 4 was not included in further analysis.

The combinations of variables that best account for each principal component are shown in Table 11. Principal component 1 most strongly represents the variables temperature, chlorophyll *a,* TSS and rainfall. Principal component 2 most strongly represents salinity (negative) and nutrients (ammonium, nitrate, orthophosphate and N:P ratios). Principal component 3 represents salinity, chlorophyll *a* (negative) and organic suspended solids (negative). Observed values for BOD_5 and SOD were analyzed for each principal component by season and study site.

Regression analysis revealed significant relationships between BOD_5 and principal components 1 ($p=0.0004$) and 3 ($p=0.0003$). Bi-plots of predicted BOD_5 values revealed numerous trends. Predicted values of BOD_5 increased with positive values of principal component one (Fig. 14). There was a strong seasonal increase from winter to summer, but site designations did not appear to cluster out. Predicted values of BOD_5 decreased with principal component 3 (Fig. 15).

Table 10. Descriptors for the Principal Components Analysis (PCA).

Number	**Eigenvalue**	**Cumulative percentage**
1	4.7693	0.2981
2	2.9042	0.4796
3	2.1077	0.6113
4	2.0501	0.7395
5	1.0002	0.8020
6	0.8292	0.8538
7	0.6591	0.8950
8	0.4656	0.9241
9	0.3757	0.9476
10	0.3418	0.9689
11	0.2641	0.9854
12	0.1054	0.9920
13	0.0660	0.9961
14	0.0330	0.9982
15	0.0166	0.9992

Table 11. Eigenvectors for PCA analysis.

Principal Component

Variable	1	2	3
Depth	0.2182	-0.1465	0.2964
Temp.	**0.4036**	-0.0328	-0.1974
L DO	-0.2527	-0.1067	0.2190
pH	-0.1865	-0.1466	0.2590
Salinity	0.1087	**-0.3887**	**0.3903**
Cond.	0.2063	**-0.3636**	**0.3185**
L Chl. *a*	**0.3423**	-0.1583	**-0.3096**
L NH4	0.0690	**0.3196**	0.2212
L NIT	-0.0047	**0.4823**	0.1866
L Phos	-0.0631	**0.3601**	0.0680
L N:P	0.0663	**0.3947**	0.2784
L TSS	**0.3099**	-0.0694	-0.1289
L OSS	0.2166	0.0077	**-0.3361**
L Rain 24	**0.3514**	0.0714	0.2041
L Rain 48	**0.3405**	0.1050	0.2148
L Rain 72	**0.3517**	0.1405	0.1588

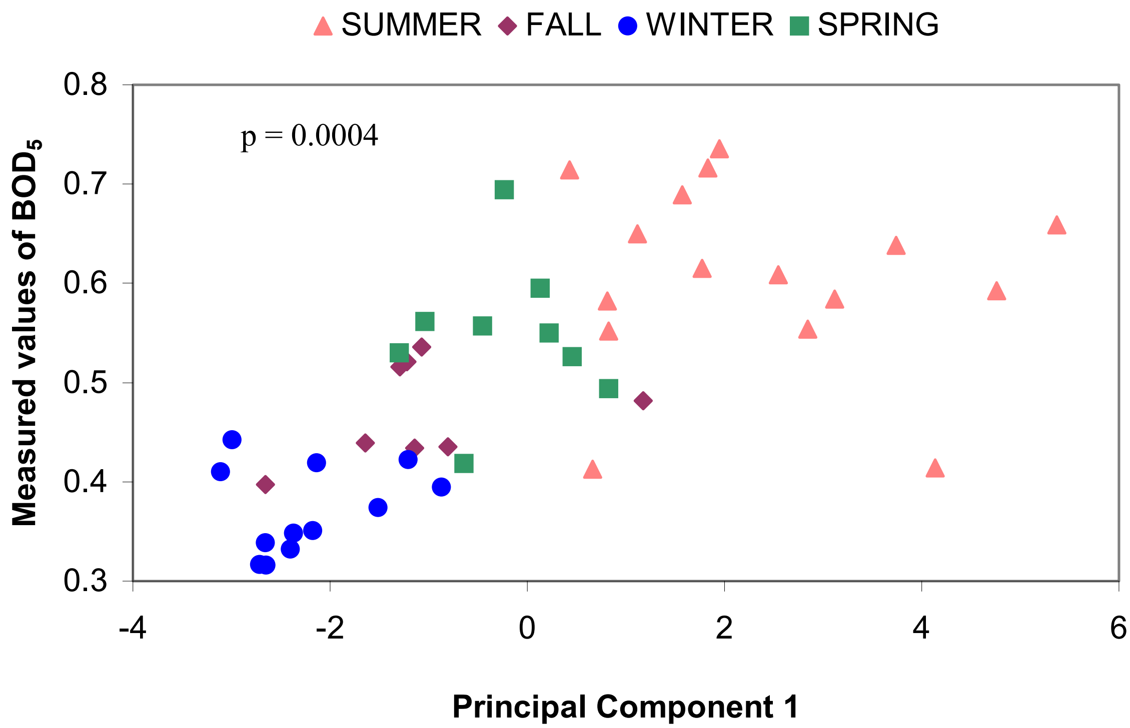

Figure 14. Measured values of BOD_5 and principal component 1 by sampling season.

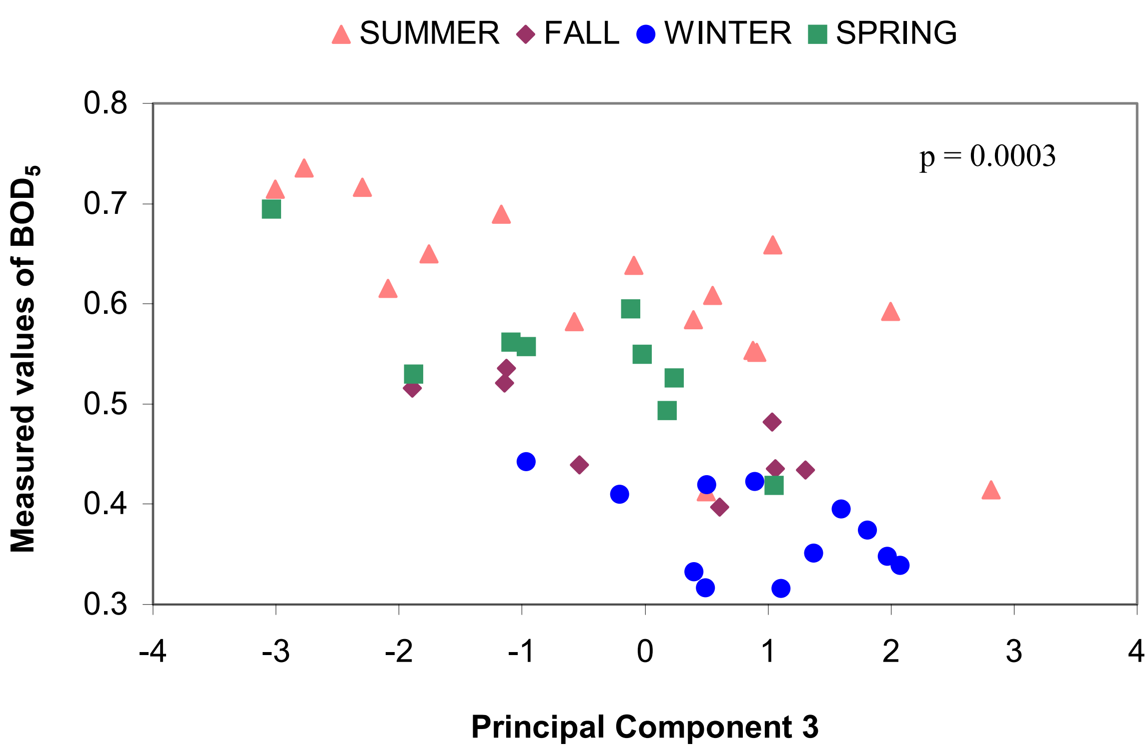

Figure 15. Measured values of BOD_5 and principal component 1 by sampling season.

Seasonal variation was apparent with predicted levels of BOD5 decreasing as summer turned to winter. BOD_5 values were lower in winter when chlorophyll *a* levels and OSS concentrations were lower. Once again, site designations did not cluster out.

Regression analysis for SOD revealed a significant effect with principal component 2 ($p=0.0447$).   The bi-plot in Figure 16 represents a strong negative trend of lower predicted values of SOD with increasing values of principal component 2, this occurred regardless of season. SOD decreases where salinity is lower and nutrients are higher (spring effect). Study sites with significantly different mean SOD rates cluster out in the bi-plot in Figure 21.   Predicted values of SOD are higher at PC-BDDS and are lower at FC-17.

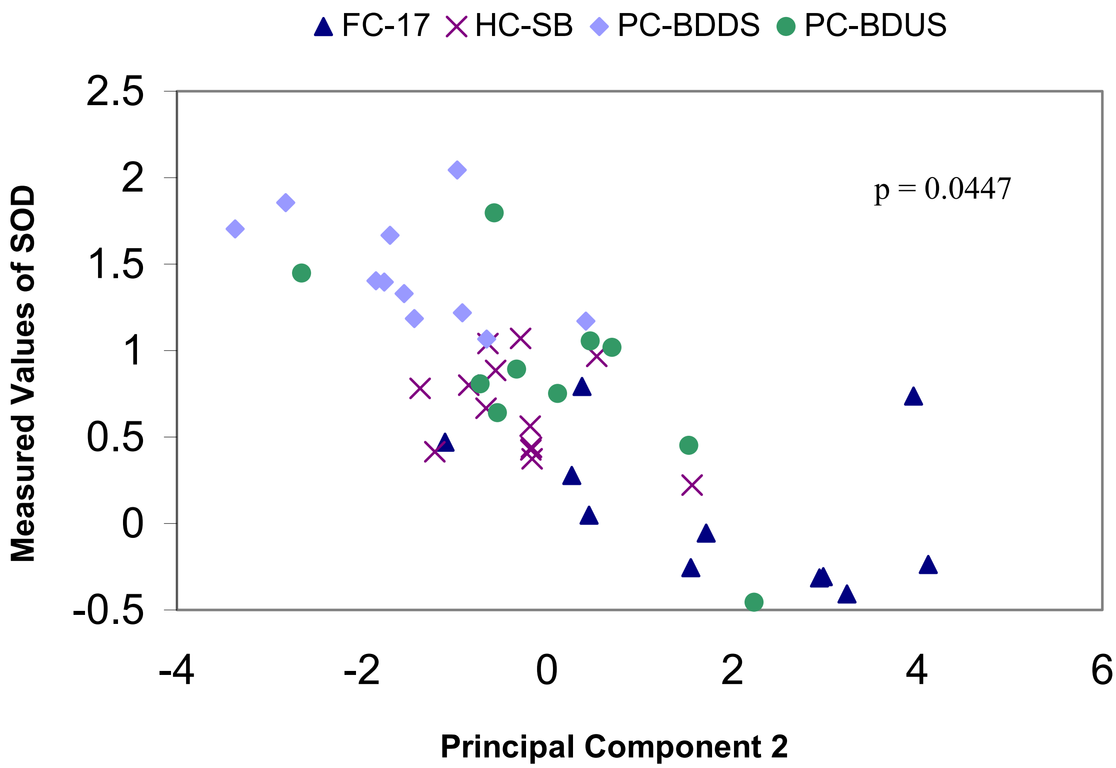

Figure 16. Measured values of SOD and principal component 2 by study site.

DISCUSSION

Water column oxygen demand (BOD_5) was not the major cause of oxygen depletion in this study. These results are consistent with Rounds and Doyle (1997), who observed that water column oxygen demand was not a significant source of oxygen consumption when measured concurrently with SOD. In the present study BOD_5 rates measured in creek water accounted for less than 31% of the oxygen loss observed using SOD chambers when SOD rates were positive. Thus, SOD is a very important contributor to dissolved oxygen deficits in local tidal creek study sites.

The magnitudes of SOD rates measured in this study are similar to what would be expected for estuarine mud/sand environments (Porcella et al., 1950), except for the negative rates seen at spring sites. Other studies (Caldwell and Doyle, 1995; Rounds and Doyle, 1997) conducted with river sediments also measured similar rates of SOD, with some higher levels reported. Most previous data did not exhibit the same amount of spatial variation in sediment oxygen demand, but a study by Mancini et al., (1950) revealed high spatial variability in SOD measurements made in Black Creek, Georgia. Seasonal variation, however, has been reported in numerous studies including those mentioned above. In this study, sites with natural springs (FC-17 and PC-BDUS) tended to have much lower SOD rates; some were even negative. This may be due to groundwater from natural springs seeping in underneath the chambers physically preventing sediment oxygen consumption.

Biochemical oxygen demand measurements reveal that spring and summertime BOD_5 rates were important oxygen sinks in local tidal creek study sites. Decreases in dissolved oxygen levels resulted from high levels of algal biological activity and subsequent labile BOD. This usually occurred in the warmer months so it also follows that DO (negative correlation) and chlorophyll *a* were correlated with temperature. Dissolved oxygen levels were higher in the colder months when there were cooler temperatures, less biological activity and less suspended sediment in the water column. Thus, we can conclude that in the summer chlorophyll *a*, temperature, TSS and thus labile biochemical oxygen demand levels were high resulting in a decrease in tidal creek dissolved oxygen measurements.

SOD was much more variable than BOD_5 indicating that SOD was an important oxygen sink that varied over time. Mean BOD_5 rates for each study site were not significantly different. Coefficients of variation for BOD_5 and SOD rates were 0.61 and 1.97 respectively. SOD rates were seasonally dependent. SOD rates were highest in the winter and spring and lowest in the fall and summer months. The high winter SOD rates contrasted with previous studies that found increases in SOD along with increasing temperatures (Seiki et al., 1994; Hu et al., 2001). Although SOD appeared to be more dominant in the winter months, there was a decrease in BOD that occurred when the water temperatures cooled and biological activity was greatly reduced.

Yearly rainfall for the 2001-2002 sampling period was 21 percent below normal. During much of this time, southeastern North Carolina experienced drought conditions. Correlations between rainfall and nutrient data indicated that runoff due to rainfall increased BOD, contributing to decreased dissolved oxygen levels in the water column. There were no significant correlations between SOD rates and rainfall when data from all sites were combined. During a wet year rates of oxygen demand may be higher, thus BOD_5 and SOD rates reported in this study should be considered conservative.

Spring sites identified in this study were previously established by Mallin et al. (1996) and Roberts (2002). Springs are not uncommon in the southeastern coastal plain. Dominant springs at FC-17 and PC-BDUS appear to have had an important influence on sediment oxygen demand rates. Sediment oxygen uptake, although still an important oxygen sink, was often low at these sites. The physical upwelling of groundwater from natural seeps may have prevented oxygen uptake from occurring in benthic chambers

Different nutrient regimes were observed at the FC-17 and PC-BDUS spring sites, and there were significant correlations between sediment oxygen demand and nutrients from the PCA analysis. Surface springs adjacent to study site FC-17 were important sources of nitrate (Mallin et al., 1996). The PC-BDUS spring had much higher ammonia levels than the Futch Creek stations in this study. Orthophosphate levels were equivalent in both springs. Nitrogen levels correlated with rainfall at FC-17 indicating that increases in N may be from runoff or groundwater. The Porters Neck Golf Course upstream of these creeks is the likely nitrogen contributor (Roberts, 2002). Nitrate levels often exceeded the North Carolina state drinking water standard of 10 mg/l. There are high inorganic nitrogen to phosphorus ratios at this site compared to others, thus the addition of orthophosphate could induce algae blooms affecting oxygen uptake at the FC-17 site (Mallin et al., 2003).

There was an overall significant correlation between SOD and organic content of bottom sediments as was hypothesized. Even sites with low organic content in bottom sediments and larger grain sizes had average SOD rates that were high enough to be important oxygen sinks, however. Ratios of total: soluble carbohydrates were greater at sandier sites than in sites with high bottom sediment organic content and smaller grain sizes. Increased soluble carbohydrates ratios may be related to the high benthic microalgal biomass associated with larger sediment grain sizes (Cahoon, 1999). Thus bioavailability of organics may be a factor influencing sediment oxygen demand as was suggested by Walker and Snodgrass, (1986).

Correlation matrices demonstrated that many variables co-varied in this data set, thus it was inappropriate to try to understand cause and effect relationships from simple pair-wise analysis alone. A principal component analysis was conducted to resolve correlation patterns in the data set. BOD_5 and SOD responded to suites of different environmental factors. BOD_5 responded to a combination of temperature, chlorophyll *a*, TSS, OSS, salinity and rainfall. SOD responded in an unexpected way to nutrients. Measured values of SOD increased with lower values of principal component 2. The (positive) nutrient values and (negative) salinity values characterize spring sites FC-17 and PC-BDUS where SOD rates are the lowest. Location may explain SOD rates rather than nutrient concentrations. Higher SOD rates occurred in the winter when there was elevated nutrient loading, but very little water column chlorophyll *a,* and thus low BOD_5 rates, to take up nutrients (There was a large amount of variability in SOD rates for this study). Principal components that were significant ($p <0.05$) in this analysis only explained 61 percent of the variability of the data set). Models for predicting BOD_5 and SOD must incorporate a suite of environmental variables. It is especially difficult to create models to predict SOD rates as numerous variables need to be taken into account.

CONCLUSIONS

Major points of this study are summarized below:

1. Mean SOD values (g O_2 m^{-2} d^{-1}) were greater than mean BOD_5 values for each month at all study sites. When monitoring aquatic ecosystem health it is important to note that BOD_5 measurements alone may not be enough to explain oxygen deficit in tidal creek systems. The large variation and greater mean SOD values for each month indicate that SOD is a large oxygen sink that may be missed if not measured directly. SOD and BOD measurements should both be considered in all comprehensive water-quality monitoring programs where dissolved oxygen levels are a problem.
2. BOD and SOD rates respond differently to numerous environmental variables. Principal component analysis reveals that a complex suite of variables must be considered in order to predict total oxygen demand.
3. BOD_5 and SOD rates were seasonally dependent. Oxygen uptake varied with numerous parameters. BOD_5 was correlated with chlorophyll *a*, TSS and OSS. These variables were all related to temperature change and thus seasonality. Thus, it is important to measure oxygen uptake rates monthly or bimonthly when monitoring aquatic systems.
4. SOD was significantly correlated with organic sediment content. Tidal creeks with high sedimentation often have high organic content in bottom sediments, which can lead to high rates of sediment oxygen demand depleting local waters of dissolved oxygen.

5. Total: soluble carbohydrate ratios indicate that the bioavailability of organic content may influence SOD rates. In this study, sites with bottom sediment characterized by larger grain sizes had the highest percentage of soluble carbohydrates indicating that even sandy sites can be important oxygen sinks.
6. Negative SOD rates occurred in areas where there are natural springs or groundwater upwelling. Groundwater was suspected to influence SOD rates in this study. Pollution in the form of excess nutrients may be carried through groundwater, thus recharge areas may be critical to tidal creek health. This concept may be key to protecting water quality. Source areas of these groundwater inputs need to be monitored. Low impact development and buffer zones should be considered priorities in these areas as well as along local tidal creek shorelines.

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